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Leland W. Briggle
Iowa State College

NOTE TO USERS

A COMPARISON OF CYTOPLASMIC-GENOTYPIC INTERACTIONS IN A GROUP OF CYTOPLASMIC MALE STERILE CORN TYPES

by

Leland W. Briggle

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INTRODUCTION

In the production of hybrid seed corn the necessity of controlled pollination on a large scale presents many problems. Crosses are produced in the conventional manner by planting two inbred lines or two single crosses side by side in an isolated field. Tassels are removed by hand, before anthesis, from the seed parent so that only hybrid seed will be borne on these plants. The process of detasseling is costly under even the most favorable conditions. Weather hazards and inadequate labor at the critical time can present further difficulties. Decided interest has arisen in the application of cytoplasmic male-sterility to the commercial production of hybrid seed corn. In the mind of the producer the optimum utilization of this genetic contribution would allow discontinuance of the detasseling procedure.

Any cytoplasmic male-sterile source, in order to be suitable for use in production of hybrids, must meet specific requirements:

1. Expression of sterility must remain stable under a wide range of environmental conditions.
2. Mode of inheritance of the male-sterile character must be simple enough to facilitate introduction into other lines.
3. Effective fertility restorer genes must be available, and easily transferred to the lines used as male parents.
4. The male-sterile character cannot have any adverse effect on yield or other agronomic characters.

It is likely that cytoplasmic male-sterility in corn occurs more frequently than had been realized. A systematic search for, and thorough testing of, each observed incidence of male-sterility may lead to isolation of types having greater value than those now being used commercially.

This investigation was undertaken to compare eight different sources of cytoplasmic male-sterility, to evaluate the commercial possibilities of each, and to compare the cytoplasmic-genotypic interactions involved.

The full potential of any source of sterility cannot be realized until an analysis of its mode of inheritance can be made. This fact prompted the preliminary genetic study on three new cytoplasmic male-sterile types which had been observed in the breeding and genetic material at Ames, Iowa. Investigations into the genetics of the other sources in the collection are either underway or have been completed by other workers.

REVIEW OF LITERATURE

In the science of genetics, nuclear inheritance has proved to be the general rule. Extranuclear inheritance seems to be the exception, although in recent years there has been an increasing number of reports of transmission of inherited characteristics in some extranuclear fashion. Several possible explanations have been presented. Aside from maternal inheritance of a transient nature, which will not be considered in this review, numerous examples suggest some type of self-reproducing autonomous element in the cytoplasm.

Cytoplasmic Inheritance of Various Characters

Cytoplasmic inheritance appears to be quite general, occurring in a number of different organisms, both plant and animal. No instance, however, has yet been reported in the vertebrates (40). Expression of several attributes is affected by some form of transmission from one generation to the next through the cytoplasm.

One type of extranuclear inheritance is involved in a series of experiments on Paramecium by Sonneborn (37, 38) and Preer (27). Certain races of Paramecium known as "killers" have a lethal effect on "sensitive" races, when the sensitives are exposed to fluids in which the killer race has lived. The character killer is governed by the presence of a dominant gene K and a specific cytoplasmic factor designated as kappa.

Lindegren (22) noted that genes control the adaptation of some yeasts to melibiose and to galactose. In experiments on adaptation to galactose with hybrids between the common bread yeast Saccharomyces cerevisiae and S. Bayanus, genes initiated production of the adaptive enzymes, but adaptation occurred only by interaction with cytoplasm of the cells with a specific substrate. That the cytoplasmic elements in yeasts are particulate is reported by Spiegelman (39); they are randomly distributed between mother and daughter cells. It is thereby possible for two cells of the same genotype to exhibit heritable differences in enzyme-forming ability.

Interracial reciprocal crosses in both the Tritici and the Avenae varieties of Puccinia graminis have produced progenies that do not have identical pathogenicity (13). Pathogenic differences displayed in F_1 persist in F_2 and F_3, while other characters not affected by the cytoplasm segregate in an expected fashion.

Resistance to infection by nodule bacteria in red clover is inherited as a simple recessive (r) combined with a cytoplasmic component (25).

Michaelis (23) reported that all major differences revealed by reciprocal crosses in Epilobium were produced by interactions between cytoplasm and nuclear genes.

In a study on Drosophila, L'Heritier (21) found that marked sensitivity to CO_2 was transmitted through the female parent only, with few exceptions. It appeared that some self-duplicating cytoplasmic particle was responsible, and in some respects resembled an infectious particle.

Cytoplasmic Inheritance of Chlorophyll Abnormalities

Early reports of non-Mendelian inheritance in plants were based on studies of certain chlorophyll abnormalities, usually expressed in the form of variegation or striping. According to Rhoades (32), Correns first described this type of heredity in Mirabilis jalapa. Transmission of the character was through the female parent only, the pollen parent having no effect. Seed from flowers borne on green branches gave only green offspring, white (pale green or yellow) branches produced only white progeny, and seed from variegated branches yielded green, white, and variegated individuals. It has been demonstrated (40) that there are self-duplicating elements outside the nucleus which have continuity from one generation to the next. These elements are the plastids which are involved in color variations of this type. Those seeds borne on a green branch would be expected to include only green plastid primordia, those from a white branch only the white plastid primordia, and seeds from a variegated branch either pale or green or a mixture of the two. Transmission of the variegated character shows evidence of a self-duplicating cytoplasmic factor, assumed to be the plastids. According to Randolph (28), Correns believed the absence of chlorophyll was due to a cytoplasmic disease which manifested itself in the plastids.

Similar situations of strictly maternal transmission have been found in Antirrhinum and Primula. In Hydrangea the inheritance of various chlorophyll patterns is likewise strictly maternal, while the transmission of white tissue in Pelargonium is biparental, but seems to fit no definite ratio (4). It is generally believed that no cytoplasm is contributed to

the zygote by the male gamete, but in the case of Pelargonium, genetical evidence seems to suggest cytoplasmic transmission to some degree through the pollen.

Karper (18) noted that variegated sorghum plants give normal green, striped, or yellow or white progeny, in accordance with the location of the parent seed in relation to the chlorophyll pattern of the panicle. He states that the nature of the progeny depends upon the nature of the maternal tissue rather than upon the genic complement.

Anderson (1) noted in a study on the same characteristic in maize that seed from specific areas on the ears gave only pale or only green seedlings. Seed at the borders of the areas usually gave rise to striped seedlings. Randolph (28) carried on a cytological investigation of Anderson's material, but did not find any difference between the cells of the green plants and the cells of normal plants of a different strain. He did report a difference in the maximum size and depth of color attained by the chloroplasts in yellowish green plants compared to those of the green plants, both types belonging to the maternal inheritance strain. There were fewer plastids in the cells of pale areas of the leaves, but a higher number of proplastids than in the cells of a green plant. Demerec (6) reported on a second case of maternal inheritance in maize, essentially the same as in Anderson's study. He likewise found that the variegation was inherited only through the female gametes, and that the male gametes had no influence on transmission or expression of the character.

Among the characters in maize which are genetically controlled is

that of iojap, expressed as a chlorophyll striping or variegation. Rhoades (31) reported that apparently the ii gene, when homozygous, is able to induce a modification in the plastids. Somatic segregation of two plastid types, those containing chlorophyll and thereby green in color and the mutated plastids which are colorless, gives the phenotypic striped pattern. Once the condition has been induced by action of the ii gene, the color types follow a strictly maternal inheritance pattern much like that reported by Anderson (1) and by Demerec (6). Rhoades (31) believed that self-duplicating bodies with non-Mendelian heredity resulted from the effect of the ii gene.

Cytoplasmic Inheritance of Male-Sterility

A number of genes conditioning male-sterility have been reported in the literature. Fewer instances are on record of a genic-cytoplasmic interaction controlling the expression of male-sterility. The cases of strictly cytoplasmic inheritance of this characteristic are rare.

As early as 1921 Bateson and Gairdner (2) found a type of male-sterility in flax. They observed male sterile plants in the F_2 generation of a cross between a procumbent race and common tall flax, but only when the procumbent race was used as the female parent. The F_1 of such a combination was normal, while the F_2 segregated three fertiles to one male-sterile. In 1927 Chittenden and Pellew (5) suggested that Bateson and Gairdner's data showed evidence of an interaction between gene and cytoplasm. They pointed out that no sterility occurred in the F_1 or F_2 of the reciprocal cross tall x procumbent. Male-sterility occurred only

when there was cytoplasmic continuity from procumbent and when the progenies were homozygous for a specific recessive gene originally brought into the cross from common tall flax. These suggestions were later confirmed by Gairdner (11), who stressed that there was a difference in the cytoplasmic constituents of the two strains, apart from the factors carried by the nucleus.

In Nicotiana East (7) reported that certain self-sterility genes from N. sanderae, when combined with cytoplasm of N. langsdorffii, produced male-sterile plants. In the reciprocal cross these factors produced male-fertile plants.

Little information is available concerning the origin of cytoplasmic differences within a species (20). Possibly a cytoplasmic mutation analogous to gene mutation occurs. If so, Lewis (20) postulates that changes in the cytoplasm probably occur more rarely than do gene mutations, and that the chance of producing an effect as marked as male-sterility is not great.

Jones and Clark (16) reported in 1943 on the mode of inheritance of male-sterility in the onion, which was found to result from an interaction between a recessive nuclear gene and a non-nuclear or cytoplasmic factor. All plants with normal cytoplasm (designated as N) produce viable pollen. All male-sterile plants possess the sterile type of cytoplasm (S). The gene for male-sterility (ms) when homozygous in plants with S cytoplasm affects pollen development. It has no effect when carried by plants with N cytoplasm. Male-sterile plants have the genotype S ms ms. Jones and Clark outlined a breeding program whereby use could be made of hybrid vigor in the commercial production of onion bulbs.

Foskett (8) reported on a second case of male-sterility in the onion. Parental pedigrees showed no indication of male-sterility. The manner of inheritance proved to be identical with that reported earlier by Jones and Clark (16). Foskett concluded that not only were the genes involved allelic, but that the cytoplasmic factors conditioning male-sterility were the same in each case. Degree of sterility in segregants of various crosses was indicative of the effect of modifiers.

Male-sterility in sugar beets was studied by Owen (26). He concluded that the inheritance of semi-male-sterility and of complete male-sterility was the result of an interaction between cytoplasm and genes. Assuming two types of cytoplasm, S for male-sterility and N for normal, and X and Z for genes involved, the genotype of the completely sterile plants is given as S$\underline{XXzz}$. S$\underline{Xxzz}$ or S$\underline{xxZz}$ represent semi-male-sterile plants, usually without viable pollen, while S$\underline{XxZz}$ plants are semi-male-sterile with some degree of viable pollen. The two factor hypothesis did not fully explain all results, suggesting that modifiers may influence the expression of the male-sterile character.

Another type of cytoplasmic male-sterility was noted in Dactylis glomerata by Myers (24). The character was conditioned by a dominant gene interacting with a specific cytoplasmic factor. Progeny from seed produced on sterile parents were sterile if quadruplex, triplex, or duplex for the sterility gene. Nulliplex plants were fertile and simplex plants either sterile or fertile. A few exceptional ratios encountered were explained by assuming the presence of modifying factors in the simplex class.

Cytoplasmic male-sterility in corn was first described by Rhoades (29),(30). His analysis indicated that transmission of the male-sterile character in this instance was through the egg cytoplasm. The nature of the pollen parent had no apparent effect on expression of the character. By means of back crossing to a series of linkage tester stocks as recurrent parents, replacement of the chromosomes in the male-sterile line with chromosomes known to be free from sterility producing factors proved to have no effect on the degree of sterility. Rhoades reported microsporogenesis to be normal, but that pollen degeneration usually occurred before the first vegetative division. In an attempt to determine if the male-sterility may have been the result of a virus infection, juice was extracted from the tassel and upper internodes of male-sterile plants. The juice was inoculated into seedlings of a normal strain, and at anthesis the plants were observed. Some were selfed and their progeny likewise examined. There was no transmission of the male-sterile character via the plant juice.

A paper published in 1948 by Josephson and Jenkins (17) presents observations on male-sterility in corn. When used as the female parent in certain crosses, the inbred line J.C.33-16 seemed to contribute some factor through the cytoplasm of the egg which conditioned male-sterility. The data also show that expression of male-sterility is influenced by contributions from the pollen parent. They concluded that at least two genetic factors in combination with the cytoplasmic contribution were necessary for the expression of male-sterility. A few other lines showed evidence of a similar cytoplasmic effect. Still other lines were of a

genotype such that expression of male-sterility was suppressed when they were used as male parents in a cross. Environmental conditions influenced the expression of this particular male-sterile type.

Gabelman (9) attempted to determine the nature of the cytoplasmic factor responsible for male-sterility. Partially fertile plants were used in a study on variability in amount of viable pollen produced among florets of a given plant. None of the variation could be attributed to gene differences so it must have been due to the presence or absence of the postulated cytoplasmic particle. When the data from an individual tassel were subjected to a 2 x N chi square test the variation among florets exceeded that expected by chance. Gabelman suggested the presence of one or more cytoplasmic particles in a non-viable pollen grain and the absence of any such particles in the normal male gametes.

Jones (14) combined a cytoplasmic component (plasmagene) of male-sterility and an independent gene (chromogene) conditioning male-sterility in the same corn plants. When both types of genes were present together there seemed to be no effect of one upon the other. In agreement with an earlier report by Josephson and Jenkins (17), Jones showed evidence that certain genotypes do have the power to overcome the male-sterile condition expressed as a result of the cytoplasmic factor. He also stated that apparently the cytoplasm has little to do with ordinary variations within a species. Such characteristics as size and maturity appear to be under complete control of the chromogenes.

A very interesting occurrence of male-sterility in maize is described by Rhoades (33). The male-sterile phenotype is induced by the action of

the iojap gene, which also is directly responsible for the inception of an irreversible mutation of plastid primordia reported on earlier (31). Convincing evidence that the same gene produces a mutation of a second and different cytoplasmic factor affecting pollen development is presented. If the chlorophyll deficient cytoplasmic factor were the same as that which causes male-sterility, only the white plants (if not lethal) would be male-sterile. One would expect the tassel branches on white sectors of striped plants to contain aborted pollen and the green sectors to produce only normal pollen. Likewise all green plants should be male-fertile. This relationship does not occur. As suggested previously by Gabelman (9), Rhoades believes the cytoplasmic factor to be particulate in nature and suggests the possibility that mitochondria may be those particles. He points out the fact that prior to this study the genetic continuity of mitochondria has never been indicated, nor has there been evidence of a fundamental difference between mitochondria and plastid primordia.

A number of different sources of cytoplasmic male-sterility in corn have recently been reported (15). Jones found that plants from these various sources differed appreciably in maintenance of sterility when crossed with the same inbred lines. These differences in degree of pollen production can well be attributed to the failure of each cytoplasmic type to be identical in nature, or to the fact that the chromogenes borne by the sterile parents are not the same. Jones proposes a procedure designated as paired progeny selection which he deems necessary for the successful transfer of cytoplasmic male-sterility to inbred lines. A cross is made with the male-sterile parent as the female and the male-fertile line to be converted is used as the pollen parent. By continual back-

crossing to the male-fertile line, the chromosomal complement of the line to be sterilized will in effect be combined with the cytoplasm of the male-sterile parent. It has been postulated (15) that some inbred lines which have been used as the fertile parent in the conversion procedure have segregated for pollen restoring genes. This resulted in the system whereby the individual plant used as the male parent in the backcross is also self-pollinated and grown alongside the progeny from the single backcrossed plant. Only completely sterile backcross progeny along with the fertile counterpart are selected for further propagation.

The first complete genetic analysis involving a cytoplasmic-genotypic interaction type of male-sterility in maize was reported on by Schwartz (36) in 1951. This particular condition of male-sterility is generally known as the "Kys" sterile. The inheritance involves an interaction among three factors. In order for sterility to be expressed a specific cytoplasm must be in combination with a dominant gene for male-sterility ($\underline{Ms}_{21}$) and a homozygous recessive gene designated as $\underline{s}^{ga}$ $\underline{s}^{ga}$ because of its association with a male gametophyte effect. The genotype of a sterile plant would be $\square$ $\underline{Ms}$ $\underline{ms}$ $\underline{s}^{ga}$ $\underline{s}^{ga}$ (the symbol $\square$ represents the male-sterile cytoplasm). The inbred line Kys, which does not act as a fertility restorer, is of the genotype O $\underline{ms}$ $\underline{ms}$ $\underline{s}^{ga}$ $\underline{s}^{ga}$ (the symbol O represents normal cytoplasm). All corn belt lines, with the exception of Kys, act as fertility restorers when crossed with "Kys" male-sterility. They are of the genotype O $\underline{Ms}$ $\underline{Ms}$ $\underline{S}^{Ga}$ $\underline{S}^{Ga}$. Due to complete selective fertilization the suppressor gene ($\underline{S}^{Ga}$) can be transmitted only when the heterozygote is used as the female parent.

In further studies on this same "Kys" sterile Bauman (3) found that plants heterozygous for the suppressor gene could be identified upon microscopic examination of the pollen grains. About 50 per cent of the pollen does not stain completely in iodine solution. Instead of a strict competition effect between $\underline{S}^{Ga}$ and $\underline{s}^{ga}$ gametes, as Schwartz (36) had reported, the pollen grains bearing the $\underline{s}^{ga}$ gene actually abort.

Rogers and Edwardson (34) evaluated the commercial possibilities of utilizing a type of cytoplasmic sterility first observed in the Texas variety, Mexican June. When this particular male-sterility is induced into certain inbred lines through a backcrossing procedure, the male-sterile character is quite stable. Crosses of male sterile Tx 203 (an inbred line converted to sterility) with various inbred lines revealed that many lines tested have no effect on the expression of sterility, since the single crosses produce no pollen. A few select lines, K55, Tx127C, and TxGJ39 carry genetic factors which restore complete fertility in the F_1 generation. Other lines tested are variable in their reaction with the sterile parent, as some partially fertile plants occur in the progenies. Based on Rogers' data, the pollinator parent of a double cross in which the Texas male-sterility is utilized should include one or two inbreds capable of restoring fertility. The alternative means of assuring the farmer ample pollen production in his corn field is the practice of blending seed produced in the conventional manner with that produced through use of male-sterile seed parents and pollinator lines not carrying restorer genes. In preliminary data listed by Rogers, sterile single crosses appear as good, or possibly better in yielding ability, than single crosses between the fertile counterpart lines.

Gabelman (10) reported that the line Minnesota A34 carried a single dominant gene which restored partial fertility to cytoplasmically male-sterile hybrids and inbreds. The source of sterility is not indicated in the abstract. F_2 data were suggestive of the presence of modifiers.

MATERIALS AND METHODS

Cytoplasmic Male-Steriles and Tester Differentials

A total of eight cytoplasmic male-steriles, each from a different source, was collected for purposes of comparison and evaluation. Three of these originated from the genetic and breeding material at Ames, Iowa.

The U.S.D.A. source, designated by a superscript S, was obtained from the Connecticut Agricultural Experiment Station. This cytoplasmic male-sterile was originally found by Jenkins, of the United States Department of Agriculture, in a cross of teopod by a linkage tester (15).

A second source was also obtained through the Connecticut Agricultural Experiment Station. This type was discovered in a South American variety by Brieger in Brazil (15). It has generally been reported that the Brazilian male-sterile is rather unstable, having shed pollen in northern areas where it has been grown. During the two seasons it has been under observation at Ames, Iowa, the male-sterile inbred line $A158^{B}$ has maintained complete sterility. The superscript B specifies the Brazilian source.

The Texas Agricultural Experiment Station, and again the Connecticut Station, furnished seed of the Texas source of male-sterility, which had been found by Mangelsdorf in the variety Mexican June (34). What is believed to be the same form of cytoplasmic sterility was observed later in closely related Texas varieties. This particular source appears under most conditions to be less variable in expression of sterility than do

the U.S.D.A. and the Brazilian types. In preliminary production of male-sterile seed stocks, the Texas source has probably been the most commonly used. In this instance the superscript T symbolizes the type of male-sterile involved.

A male-sterile single cross, J.C.33-16 x Mo2RF, from the Kentucky Agricultural Experiment Station was included in the collection of male-steriles. The female parent, inbred line J.C.33-16, has been described in the literature review. The cross, J.C.33-16 x K63, was likewise obtained but proved to be fertile when grown at Ames, Iowa, and was eliminated from the tests.

The cytoplasmic male-sterile commonly known as "Kys" appeared to be quite different from the other types under observation. An account on the manner of inheritance has been given in the literature review. Seed of the male-sterile stock was obtained from Schwartz. Previous reports have indicated that expression of sterility does not seem to be influenced by environment (36). This characteristic would be particularly desirable in a type of sterility which could be converted to commercial use. In the case of the Kys source, the complexity arising from the action of the male gametophyte factor presents a problem in the transfer of the male-sterility to another line. In the material grown at Ames over a period of two seasons, none of the Kys male-sterile plants showed any indication of anther exsertion or of pollen shedding.

A condition of incomplete male-sterility existing in the single cross M1984 x M14 was noticed in the pollinating nursery at Ames, Iowa. Practically no pollen was shed by tassels of this combination, but when M1984 was used as the male parent in the same cross, the tassels of the hybrid

plants shed normally. Such a marked difference between reciprocal crosses involving M1984 and M14 suggested a cytoplasmic effect, and the incompletely male-sterile single cross was included as one of the eight sources of cytoplasmic male-sterility.

Male-sterile plants were observed by Sprague in a genetic stock of the genotype Vg sy j/sy y_{16}. Approximately 50 per cent of the plants bore the dominant gene, vestigial glume (Vg), thus being functionally male-sterile. The very short glumes afford no protection for the anthers and they dry up before time of pollen shedding. The remaining plants in the population had normal tassels, except for the fact that no anthers were exserted. These plants lacking the Vg gene exhibited a type of complete male-sterility, and when pollinated by certain inbred lines or genetic stocks produced male-sterile progeny. Absence of partially fertile plants indicated potential use of this type of sterility. It represents the second of the Iowa Agricultural Experiment Station contributions to the cytoplasmic male-sterile collection. This particular male-sterile is designated by use of the superscript Vg, and will be referred to as such in the following sections of this paper. The name is derived from the genetic stock it originated from and has no reference to the effect of the Vg gene.

In 1951 an isolated block of Reid Yellow Dent was grown at Ames. Open pollinated ears were saved from a total of 70 plants which had proved to be male-sterile. Seed from each ear was planted in a row the following season in a search for possible cytoplasmic male-sterility. The progeny from one ear was sterile, with the exception of two partially fertile plants. Two other ear rows had a few male-sterile plants interspersed

with fertile plants. Only the first progeny row mentioned showed promise of being a type of cytoplasmic male-sterility, so was included in the collection to be tested. Based on the previous system of symbols, the Reid cytoplasmic male-sterile is designated by a superscript R.

Conventional genetic tests cannot be applied to male-sterile stocks. An indirect method has to be adopted to test differences between sources of sterility, such as use of a series of tester lines serving as fertility differentials. An attempt was made, within limits of information available, to include in a series of tester differentials one fertility restoring inbred line for each of the eight cytoplasmic male-sterile sources, and one line which had proved not be a fertility restorer. At the time the series was made up no test crosses had been made on the three sources from the Iowa breeding and genetic material. In the case of the other five cytoplasmic male-steriles, some of the tester lines chosen were known to be fertility restorers or non-restorers to more than one. Inbred lines included in the tester differential series are K63, Tx127C, TxGJ39, K64, Ky39, Ky21, K55, M14, Mo2RF, Kys, and a multiple tester genetic stock originally obtained from Mangesldorf.

Experimental Methods

Effects of date of planting

Environment seems to affect expression of cytoplasmic male-sterility in most cases reported to date. In order to compare the eight previously described sources as to some degree of environmental influence, each type of cytoplasmic male-sterile was planted at three different dates. Single

crosses involving each of the eight sterility sources with the complete series of tester differentials were included in the date of planting experiment. The first planting was made on May 9, the second on May 26, and the third on June 11. This test was grown only in 1953. Because of limited land available for the amount of material to be grown, only a single 25 plant row represented each entry at one planting date.

When all plants within a row were in full silk, the tassels were classified individually, and the data recorded. Each row was checked at least once from two to five days after the first observation. Some required much closer scrutiny than others, so that a number of checks were made on the more variable rows. Tassels were classified on the basis of anther exsertion and degree of pollen shedding, as indicated in Table 1.

The data from planting date one were compared with planting date two, date two with date three, and date one with date three for each entry in the experiment. A test of independence of effects due to dates was made for each entry by computing chi square for an r x 2 table (12), the value of r depending upon the number of classes occurring in the two dates compared.

Relic heterozygosity

Persistence of heterozygosity in long-time inbred lines is a somewhat controversial issue. This problem arises in the conversion of some inbred lines into sterile counterparts. It was felt that an experiment involving two long-time inbred lines, Hy and C.I.7, which had been reported as apparently segregating for fertility restoring genes (34), might prove

Table 1. Description of classes used for the classification of sterile, partially fertile, and fertile tassels in the date of planting experiment

Class	Per cent of tassel area bearing exserted anthers with no pores	Per cent of tassel area bearing exserted anthers with visible pores
0	0	0
1	trace	0
1f		trace
2	25	0
2f		25
3	50	0
3f		50
4	75	0
4f		75
5	100	0
5f		100
5pf	Approximately 50	Approximately 50

worthwhile. Seed lots of line Hy and of line C.I.7 were obtained from five different sources, as indicated in Table 2. Each line has been maintained for a number of years under different environmental conditions and probably under separate selection procedures at each location. Progeny from some seed lots of one line showed marked phenotypic differences from

Table 2. Sources of seed lots of inbred line Hy and of inbred line C.I.7

Lot number	Source of Hy	Source of C.I.7
1	U.S.D.A. Exp. Sta., Beltsville	U.S.D.A. Exp. Sta., Beltsville
2	Illinois Agr. Exp. Sta.	Illinois Agr. Exp. Sta.
3	Kansas Agr. Exp. Sta.	Kansas Agr. Exp. Sta.
4	Kentucky Agr. Exp. Sta.	Kentucky Agr. Exp. Sta.
5	Missouri Agr. Exp. Sta.	Missouri Agr. Exp. Sta.

progeny of other seed lots of the same line. Differences noted in line Hy were of plant type, tassel type, plant height, and maturity dates. Lots of line C.I.7 were less variable, however, there were differences in plant height, susceptibility to top firing, and in maturity dates. The different seed lots, possibly representing sub-lines, certainly differ to some degree in genotype.

Original plans called for individual plant pollinations as a means for testing the genotype of Hy and C.I.7 plants, in respect to fertility restoring genes. An individual Hy plant was to be used as the pollen parent in a cross to three sterile lines, temporarily designated as line

A^S, line A^T, and line B^T. This procedure would test the individual Hy plant against a common inbred line background with two different sources of sterility, and against two different inbred lines carrying the same cytoplasmic male-sterility type. A total of ten or more plants from each seed lot of line Hy was to have been tested in this manner in 1952. The same procedure was outlined for line C.I.7 plants. Due to a combination of poor stand and dissimilarity of flowering time, the proposed plan had to be abandoned. Individual Hy and C.I.7 plants were crossed to different sterile sources and in some cases to the same source in different lines, but only male-sterile plants which had not been selected for use in the male-sterile x tester differential series were available. A number of rows of Texas cytoplasmic male-sterile single crosses were used because of the shortage of male-sterile inbred lines. The test crosses made were grown in ear rows in 1953, and data taken on degree of sterility of the progeny. The originally proposed plan for making test crosses of individual plants from each seed lot of Hy and C.I.7 was carried out in 1953, but the material will not be grown before 1954.

Morphological study of anther and pollen development in an incompletely male-sterile single cross and in the fertile reciprocal cross

The F_1 generation of M1984 x M14 is incompletely male-sterile; all plants are uniform in expression of sterility. The reciprocal cross M14 x M1984 is fully fertile.

Tassels of each cross were sampled at the time of meiosis, and at four-day intervals thereafter until time of anthesis. A total of five tassel samples were taken from each of 12 plants of M1984 x M14, and a

total of five were likewise taken from each of 12 plants of M14 x M1984. The same plant was sampled at each of the five different dates by removal of one basal tassel branch. The tassel branch was immediately immersed in 70 per cent ethyl alcohol and stored until a morphological investigation could be made. At time of meiosis larger sections of tassel were removed from five plants of M1984 x M14 which were not involved in the periodic sampling procedure. These tassel branches were immersed for a period of 24 hours in an acetic acid-alcohol killing solution and then transferred to 70 per cent alcohol until a microscopic examination of meiotic configurations could be made.

Five spikelets were selected at random from the same general area of each preserved tassel branch and the following measurements taken: glume length, anther length and anther width of the pedicellate floret and of the sessile floret, and pollen grain diameter. Measurements were made through use of a microscope and a binocular equipped with an eyepiece micrometer disk which had been calibrated by means of a stage micrometer (35). Other data recorded were stages of development of microsporocytes or pollen grains and per cent of pollen grains which stain in iodine solution.

The statistical design adopted for the morphological study was that of an n-fold hierarchal classification (19). Analyses of variance were computed for glume length, anther length in the pedicellate and in the sessile florets, anther diameter in the pedicellate and in the sessile florets, diameter of normal pollen grains for the last three sampling dates, and pollen diameter of those grains which stain with iodine solution and those which do not stain in the cross M1984 x M14.

Genetic analyses

Preliminary genetic studies were undertaken on the three sources of cytoplasmic male-sterility originating from the Iowa Agricultural Experiment Station. In 1952 a small F_2 population of M1984 x M14 was grown and classified. Completely sterile plants and normal fertile plants occurred, as well as a number of intermediate types extremely variable in degree of fertility. The classification system used was unsatisfactory, so a much larger F_2 population was grown the following year. The F_2 generation of the reciprocal cross M14 x M1984 was also grown both years. The different crosses made in 1952 and grown and classified in 1953 are given in Table 3.

Table 3. Segregating generations involving combinations of inbred lines M1984 and M14, grown and classified in 1953

Backcrosses to F_1	F_2 generations
(M1984 x M14) x M14[a]	M1984 x M14[a]
(M1984 x M14) x M1984[a]	M14 x M1984
M1984 x (M1984 x M14)[a]	
M1984 x (M14 x M1984)	
M14 x (M1984 x M14)	

[a]Planted at two different dates to test influence of environment on expression of sterility.

In an attempt to study the mode of inheritance of the Vg and of the Reid male-steriles, six inbred lines (included in the tester differential series) were selected which were believed to be possible restorers of fertility. It was planned to make F_2 generations and backcrosses to both parents for each single cross which proved to be fertile. The six lines selected were TxGJ39, K64, Ky39, Ky21, K55, and the multiple tester

genetic stock. The F_1 crosses involving both the Vg and the Reid sterile types were made in 1952 and grown in the greenhouse during the winter of 1952-53. Fertile F_1 plants were selfed and backcrossed to each parent, and the F_2 and backcross populations grown in 1953. The F_2 generations of 4632^{Vg} x Ky 21, 4632^{Vg} x Ky 39, and of 4620^{R} x Ky 21 were grown at two different planting dates so that observations might be made on degree of

Table 4. Description of nine classes used in the classification of genetic populations segregating for male-sterility

Class designation	Class description[a]
0	No anthers exserted; no pollen shed
1	Trace anthers exserted; no pollen shed
2	25 per cent anthers exserted; no (or trace) pollen shed
3	50 " " " " ; " " " " "
4	75 " " " " ; " " " " "
4pf	75 " " " " ; up to 25 per cent pollen shed
5	100 " " " " ; no (or trace) pollen shed
5pf	100 " " " " ; up to 75 per cent pollen shed
5f	100 " " " " ; up to 100 per cent pollen shed

[a]Per cent given refers to fraction of total tassel area.

environmental influence on expression of sterility. All segregating generations were classified for male-sterility into a maximum of nine classes. Description of the different classes is given in Table 4. The same system of classification was used for the segregating generations of the M1984 and M14 combinations.

EXPERIMENTAL RESULTS

Comparison of Cytoplasmic Male-Sterile Sources

Individual plants of the F_1 crosses involving each source of cytoplasmic male-sterility with each line of the tester differential series were classified as to degree of fertility. The data are presented in summary form in Table 5.

The three lines converted to the U.S.D.A. source of male-sterility reacted alike when crossed with ten of the tester lines, except for minor variation in degree of expression of sterility. The crosses involving the multiple tester stock as male parent with A^S and $A158^S$ segregated for sterility, suggesting the fact that the multiple tester may have been heterozygous for the restorer gene or genes. There is also the possibility that the male-sterile parent may have been heterozygous for a modifier gene or genes which when complemented by the male parent genotype resulted in sterility. Within limits of the test crosses made, there seems to be no difference between the U.S.D.A. and the Brazilian sources of cytoplasmic male-sterility.

The Texas source, occurring in the test in the background of three separate lines, is different from all other sterile types in reaction with the tester differentials. Lines K63 and K55 restore complete fertility when used as the male parent with the Texas source, but in crosses with the other steriles, except for the Kys type and probably J.C.33-16, only male-sterile progeny occur. Inbred lines Tx127C and TxGJ39 restore

Table 5. Degree of fertility of single crosses between cytoplasmic male-sterile sources and the series of tester differential lines

Source of sterility[a]	Tester differential										Multiple tester
	K63	Tx127C	TxGJ39	K64	Ky39	Ky21	K55	M14	Mo2RF	Kys	
A^S	-	-	Sw	-	F	F	S	S	-	S	S&F
$WF9^S$	S[b]	-	Sw	S	F	F	S	S	S	S	S
$A158^S$	S	S	Sw	S	F	F	S	S	S	S	S&F
$A158^B$	-	-	Sw	-	-	F	S	S	S	S	S&F
$A158^T$	-	F	-	PF	-	F	F	PF	-	S	S&PF
$Tx203^T$	F	F	F	PF	S	F	F	PF	S	S	-
$Tx61M^T$	F	F	F	PF	S	F	F	PF	S	S	-
J.C.33-16 x Mo2RF	-	S&F	S&F	S&F	S&F	F	S&F	S&F	S	S&F	S&F
$\frac{Msmss}{}^{ga}{}_{g}{}^{ga}$ (Kys)	F	F	F	F	F	F	F	F	F	-	-
M1984 x M14	S	S&F	Sw&F	S	F	F	S	S	S	S&F	S&F
4652^{Vg}	S	S	Sw	S	F	F	S	S	S	S	S&F
4620^R	S	-	PF	S	F	F	S	S	S	S	F

[a]4652 and 4620 are 1952 row numbers of male-sterile stocks.

[b]S-sterile; Sw-sterile, anthers exserted over entire tassel; F-fertile; PF-partially fertile, more than trace of pollen shed; S&F-sterile and fertile plants in the same progeny.

complete fertility to only the Texas and Kys forms of male-sterility. The reaction of K64 and M14 with the Texas source is different from that of any other form of male-sterility in the collection, in that the F_1 plants are partially fertile. The only sterility source producing male-sterile F_1 plants when crossed with line Ky39 is the Texas type. Only one cross was obtained with the multiple tester, but it appears again that the male parent may have been heterozygous at the locus or loci concerned, or that the male-sterile parent may have been heterozygous for modifiers. The multiple tester stock does not restore complete fertility to any of the F_1 plants deriving the cytoplasmic component from the Texas source.

The J.C.33-16 x Mo2RF cross is difficult to compare with the other sources, many of which are inbred lines. It differs from the single cross M1984 x M14 in reaction with K64, Ky 39, K55, M14, and in degree of expression of sterility with TxGJ39. Some fertile plants occur in all three-way crosses with the tester lines except in the case of Mo2RF, which is a backcross. It is possible that J.C.33-16 crossed with each of the testers would produce fertile F_1 plants. If that be true, this source differs from the other types.

The Kys source is fertile in combination with any of the tester differential lines. This is not the case when any other sterility source is considered.

The single cross M1984 x M14 in most combinations with the tester lines seems to be similar to the U.S.D.A. and Vg sources. However, there are fertile plants in the progenies when Tx127C, TxGJ39, and Kys are used as male parents in the three-way cross, and M1984 x TxGJ39 or Kys produces

fertile progeny. When M14 is used as the male parent in a single cross involving any other sterility source, the progenies are fertile, sterile, or partially fertile. The F_1 plants of the single cross M1984 x M14 are incompletely sterile, a condition quite different from even the partially fertile plants of the Texas source x M14.

As determined by the tester lines used, the Vg source seems to be similar to that of the U.S.D.A. and the Brazilian types, and not greatly unlike the Reid source.

The cytoplasmic male-sterility originating from Reid Yellow Dent appears to a degree to be unlike any of the other sources in that TxGJ39 restores partial fertility when used as the male parent in the F_1 cross. This could be attributed also to a specific modifier background of 4620^R. When the multiple tester stock was crossed with the Reid male-sterile, complete fertility was restored. The plant used as male in this cross may have been a different genotype at the locus or loci concerned with male-sterility from that of the plants used in crossing with other male-sterile types.

Table 5 indicates that the three lines converted to the U.S.D.A. form of male-sterility are alike in reaction with the tester differential lines. The same comparison can be made among the three lines which derive their cytoplasmic contribution from the Texas source. In Table 6 the more detailed data presented indicate that in some cases there is variability in degree of expression of male-sterility among crosses involving different lines, bearing the same source of sterility, with a common tester. Each cross was planted at three dates, the first being May 9, the second

Table 6. Mean fertility scores for crosses of three lines converted to the U.S.D.A. source of male-sterility and for crosses of three lines converted to the Texas source with the series of fertility differential tester lines, excluding the multiple tester, at each of three planting dates[a]

Male-sterile lines	Planting dates	Tester differentials									
		K63	Tx127C	TxGJ39	K64	Ky39	Ky21	K55	M14	Mo2RF	Kys
A^S	1	-	-	5.0	-	5.0F	5.0F	0.1	0.0	-	2.4
	2	-	-	5.0	-	5.0F	5.0F	1.2	0.0	-	3.6
	3	-	-	5.0	-	5.0F	5.0F	1.1	0.3	-	3.8
$WF9^S$	1	0.0	-	5.0	0.0	5.0F	5.0F	2.9	0.0	0.6	2.3
	2	0.0	-	5.0	0.0	5.0F	5.0F	3.2	0.0	1.4	2.4
	3	0.0	-	5.0PF	0.0	5.0F	5.0F	3.4	0.0	1.2	2.4
$A158^S$	1	0.0	3.0	5.0	0.5	5.0F	5.0f	0.0	0.3	0.4	1.1
	2	0.0	2.5	5.0	1.8	5.0F	5.0F	1.7	1.1	0.7	1.4
	3	0.0	2.0	5.0PF	3.7	5.0F	5.0F	2.1	1.4	0.8	-
$A158^T$	1	-	5.0F	-	1.6PF	5.0F	5.0F	5.0F	-	-	0.0
	2	-	5.0F	-	1.9S&PF	5.0F	5.0F	5.0F	-	-	0.0
	3	-	5.0F	-	1.7S&PF	5.0F	5.0F	5.0F	-	-	0.0
$Tx203^T$	1	5.0F	5.0F	5.0F	3.5PF	5.0F	5.0F	5.0F	2.7PF	0.0	0.0
	2	5.0F	5.0F	5.0F	2.6S&PF	5.0F	5.0F	5.0F	2.5	0.0	0.0
	3	5.0F	5.0F	5.0F	2.0S&PF	5.0F	5.0F	5.0F	1.0	0.0	0.0
$Tx61M^T$	1	5.0F	5.0F	5.0F	0.9S&PF	5.0F	5.0F	5.0F	4.3PF	0.0	0.0
	2	5.0F	5.0F	5.0F	0.4S&PF	5.0F	5.0F	5.0F	3.1	0.0	0.0
	3	5.0F	5.0F	5.0F	1.3S&PF	5.0F	5.0F	5.0F	2.1	0.0	0.0

[a]0-no anthers exserted; 1-trace anthers exserted; 2-anthers exserted on 25 per cent of tassel area; 3-anthers exserted on 50 per cent of tassel area; 4-anthers exserted on 75 per cent of tassel area; 5-anthers exserted over entire tassel; F-fertile; PF-partially fertile; S&PF-sterile and partially fertile plants in the same progeny.

May 26, and the third date June 11. Hereafter, these planting dates will be referred to as date 1, date 2, and date 3, respectively.

Table 6 indicates that among the three lines converted to the U.S.D.A. form of male-sterility there is more variability in expression of sterility in the F_1 crosses than among the three lines converted to the Texas type of male-sterility in their respective F_1 crosses. Crosses between TxGJ39 and three lines bearing the U.S.D.A. male-sterility gave similar fertility-sterility responses over the first two dates of planting (Table 7). However, for the third date of planting, the response of lines $WF9^S$ and $A158^S$ differed significantly from that of A^S. This difference in behavior suggests that the nuclear-cytoplasmic interaction systems of the three lines were not affected similarly by changes in environment. No difference among the three lines was encountered in crosses with K63, Ky39, or Ky21, but in Table 7 significant chi square values for tests of independence between crosses with K64 at each of the three dates indicates that $WF9^S$ and $A158^S$ are unlike in expression of male-sterility. Environment had no visual effect on $WF9^S$, but anther exsertion increased in $A158^S$ from date 1 through dates 2 and 3. A different background in the two lines concerned appears to provide a possible explanation.

Several more comparisons are listed in Table 7 among crosses which did not give identical values for mean scores in Table 6. Some chi square values are not significant, for example A^S x K55 vs $A158^S$ x K55 at each of the three dates do not represent statistically different populations. A^S and $WF9^S$ are different at each of the three dates when crossed with K55, as is the case when $WF9^S$ and $A158^S$ are compared when crossed with K55. In

Table 7. Chi square values for tests of independence between two lines converted to the same male-sterility type (U.S.D.A.) crossed onto a common tester

Comparison	X^2	df	P	Means of scores compared[a]
A^S x TxGJ39 date 3 vs $WF9^S$ x TxGJ39 date 3	25.9	1	<.01	5.0-5.0PF
" x " " 3 vs $A158^S$ x " " 3	27.0	1	<.01	5.0-5.0PF
$WF9^S$ x K64 date 1 vs $A158^S$ x K64 date 1	9.0	2	>.01	0.0-0.5
" x " " 2 vs " x " " 2	21.1	5	<.01	0.0-1.8
" x " " 3 vs " x " " 3	32.0	4	<.01	0.0-3.7
A^S x K55 date 1 vs $WF9^S$ x K55 date 1	21.9	4	<.01	0.1-2.9
" x " " 2 vs " x " " 2	19.8	4	<.01	1.2-3.2
" x " " 3 vs " x " " 3	27.6	4	<.01	1.1-3.4
A^S x K55 date 1 vs $A158^S$ x K55 date 1	3.2	1	>.10	0.1-0.0
" x " " 2 vs " x " " 2	1.7	3	<.70	1.2-1.7
" x " " 3 vs " x " " 3	1.6	4	>.80	1.1-2.1
$WF9^S$ x K55 date 1 vs $A158^S$ x K55 date 1	33.0	4	<.01	2.9-0.0
" x " " 2 vs " x " " 2	10.1	4	<.05	3.2-1.7
" x " " 3 vs " x " " 3	14.9	4	<.01	3.4-2.1
A^S x M14 date 1 vs $A158^S$ x M14 date 1	4.3	2	>.10	0.0-0.3
" x " " 2 vs " x " " 2	29.7	2	<.01	0.0-1.1
" x " " 3 vs " x " " 3	29.4	3	<.01	0.3-1.4
A^S x M14 date 3 vs $WF9^S$ x M14 date 3	2.7	3	<.50	0.3-0.0
$WF9^S$ x M14 date 1 vs $A158^S$ x M14 date 1	3.3	2	<.20	0.0-0.3
" x " " 2 vs " x " " 2	25.8	2	<.01	0.0-1.1
" x " " 3 vs " x " " 3	34.0	3	<.01	0.0-1.4
$WF9^S$ x Mo2RF date 1 vs $A158^S$ x Mo2RF date 1	1.5	2	<.50	0.6-0.4
" x " " 2 vs " x " " 2	5.9	3	>.10	1.4-0.7
" x " " 3 vs " x " " 3	4.0	2	>.10	1.2-0.8
A^S x Kys date 1 vs $WF9^S$ x Kys date 1	3.0	5	.70	2.4-2.3
" x " " 2 vs " x " " 2	6.4	4	<.20	3.6-2.4
" x " " 3 vs " x " " 3	11.2	4	>.02	3.8-2.4
A^S x Kys date 1 vs $A158^S$ x Kys date 1	5.4	5	>.30	2.4-1.1
" x " " 2 vs " x " " 2	16.7	5	<.01	3.6-1.4
$WF9^S$ x Kys date 1 vs $A158^S$ x Kys date 1	8.2	4	<.10	2.3-1.1
" x " " 2 vs " x " " 2	6.7	4	<.20	2.4-1.4

[a]Means of scores taken from Table 6.

examples such as these where all three dates differ, it seems that an explanation based on modifier background would be more sound than one based on the effect of fluctuating environmental conditions such as differences in temperature and (or) humidity at time of flowering.

When the same three lines were crossed with M14, A^S compared with $A158^S$ at date 1 gave a P value of .10, while P values of less than .01 were obtained at the other two dates. Much the same situation occurs when comparing $WF9^S$ and $A158^S$. A^S and $WF9^S$ are alike, but each differs markedly from $A158^S$ at the last two dates.

$WF9^S$ and $A158^S$ do not differ significantly when crossed with Mo2RF at any of the three dates.

Chi square for A^S x Kys compared to $WF9^S$ x Kys is significant only at date 3. A^S and $A158^S$ when crossed with Kys differ at date 2, while $WF9^S$ and $A158^S$ are alike in reaction with Kys at the first two dates. Unfortunately there was no third planting of $A158^S$ x Kys because of shortage of seed.

The three lines converted to the Texas form of male-sterility show variation in expression of male-sterility only in the tester crosses involving K64 and M14 (Table 6). When $A158^T$ and $Tx203^T$ are crossed to K64 the first two planting dates listed in Table 8 show a difference in degree of male-sterility. When $A158^T$ x K64 and $Tx61M^T$ x K64 are compared, the P values for the first and second planting dates were $>.05$ and $<.01$ respectively. Likewise $Tx203^T$ x K64 and $Tx61M^T$ x K64 differ at the first two planting dates. All three comparisons are similar for date 3. Since chi square values for all first ($A158^T$ x K64 vs $Tx61M^T$ x K64 was barely non-significant) and all second planting dates were significant, one can assume

that the male-sterile lines were different. Although no P values for the third date comparisons indicated real differences, the values range from .70 to .10.

Only two lines can be compared in crosses to M14, but here environmental effect seems quite marked. The first planting date of both $Tx203^T$

Table 8. Chi square values for tests of independence between two lines converted to the same male-sterility type (Texas) crossed onto a common tester

Comparison	X^2	df	P	Means of scores compared[a]
$A158^T$ x K64 date 1 vs $Tx203^T$ x K64 date 1	10.2	3	<.02	1.6-3.5
" x " " 2 vs " x " " 2	11.5	5	<.05	1.9-2.6
" x " " 3 vs " x " " 3	2.6	4	<.70	1.7-2.0
$A158^T$ x K64 date 1 vs $Tx61M^T$ x K64 date 1	7.2	3	>.05	1.6-0.9
" x " " 2 vs " x " " 2	14.7	4	<.01	1.9-0.4
" x " " 3 vs " x " " 3	2.1	3	>.50	1.7-1.3
$Tx203^T$ x K64 date 1 vs $Tx61M^T$ x K64 date 1	25.4	4	<.01	3.5-0.9
" x " " 2 vs " x " " 2	29.2	4	<.01	2.6-0.4
" x " " 3 vs " x " " 3	6.9	4	>.10	2.0-1.3
$Tx203^T$ x M14 date 1 vs $Tx61M^T$ x M14 date 1	10.0	4	<.05	2.7-4.3
" x " " 2 vs " x " " 2	4.0	5	>.50	2.5-3.1
" x " " 3 vs " x " " 3	10.7	5	>.05	1.0-2.1

[a]Means of scores taken from Table 6.

x M14 and $Tx61M^T$ x M14 show a high mean score, the second date an intermediate mean score, and the third date a lower mean score. A difference between $Tx203^T$ and $Tx61M^T$ is indicated at date 1 but not at date 2. Date 3 is questionable as it is barely non-significant.

Environmental Influence on Expression of Male-Sterility

Each of the sources of cytoplasmic male-sterility, with the exception of the male-sterile single cross J.C.33-16 x Mo2RF, was planted at the three different dates. Three different inbred lines converted to the U.S.D.A. cytoplasmic male-sterility were included, but the other sources were represented by a single entry each. Chi square values, computed in tests of independence between effects on expression of male-sterility by dates of planting, are listed in Table 9, as are mean scores based on the classification system for area of anther exsertion on the tassels. Table 9 indicates no environmental influence, due to varying dates of planting, on expression of male sterility in A^S, $WF9^S$, $A158^S$, $A158^B$, $A158^T$, and the Kys type, all of which can be assumed to be reasonably homozygous. The Vg source likewise is unaffected by different dates of planting but is not a homozygous stock. The difference between either of the first two dates of planting of M1984 x M14 and the third date can be attributed entirely to environmental influence. The cross between two long-time inbred lines, M1984 and M14, is represented at each of the three planting dates by the same genotype and the same cytoplasmic component. The Reid source of cytoplasmic male-sterility is somewhat variable in expression of sterility when dates of planting are compared but the chi square values are well within the limits expected due to chance fluctuations. The 4620^R stock is very likely a highly heterozygous population.

Tables 10 through 16 list the chi square values for tests of independence between effects on expression of male-sterility through dates of planting of crosses involving the sources of cytoplasmic male-sterility

Table 9. Chi square values computed from tests of independence between effects due to planting dates for each source of cytoplasmic male-sterility, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$WF9^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^B$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^T$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
M1984 x M14	1	5.0	1-2	0.0		
	2	5.0	2-3	46.0	1	<.01
	3	5.0PF	1-3	47.0	1	<.01
4652^{Vg} x *sy* gl16	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
4620^R	1	1.1	1-2	0.1	1	<.80
	2	0.9	2-3	0.2	1	<.70
	3	0.7	1-3	0.6	1	<.50
Ms ms s^{ga} s^{ga}("Kys")	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		

[a]0-no anthers exserted; 1-trace anthers exserted; 2-anthers exserted on 25 per cent of tassel area; 3-anthers exserted on 50 per cent of tassel area; 4-anthers exserted on 75 per cent of tassel area; 5-anthers exserted over entire tassel; PF-all plants partially fertile.

and several of the tester differential lines. The Kys source is omitted because all tester lines restored complete fertility in the F_1 generation. The tester line Ky 21 is not included because it restores fertility to all sources of cytoplasmic male-sterility tested. Ky39 is likewise omitted because all crosses were fertile except the three-way involving J.C.33-16 x Mo2RF and the F_1 crosses involving the Texas type of male-sterility, but dates of planting had no measurable effect. Another tester, inbred line Kys, when crossed to each male-sterile source was not significantly affected in degree of expression of male-sterility at different planting dates with the exception of one instance which will be pointed out later. Several of the F_1 populations involving the multiple tester stock as male parent were segregating for sterility and fertility. These two testers, Kys and the multiple tester stock, are not listed in the following tables. The mean score based on percentage of anther exsertion over the entire tassel is included for each date of planting in the tables. The mean scores were all computed through the procedure outlined at the end of Table 9.

As shown in Table 10 line K63 can apparently be effectively sterilized by the U.S.D.A. source, but it restores complete fertility to the Texas form of male-sterility. The three sources originating at Ames are variable in reaction with K63 at the three planting dates. Dates 1 and 3, and 2 and 3 differed in the 3-way cross (M1984 x M14) x K63. The data suggest environmental influence on expression of male-sterility.

K63 crossed with the Vg source does show some anther exsertion, but this characteristic was not appreciably affected through changing the date of planting.

Table 10. Chi square values for tests of independence between date of planting effects on crosses involving five sources of cytoplasmic male-sterility with K63, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S		--	--	--	--	--
$WF9^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^B$		--	--	--	--	--
$A158^T$		--	--	--	---	--
$Tx203^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
$Tx61M^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
J.C.33-16 x Mo2RF		--	--	--	--	--
M1984 x M14	1	1.1	1-2	3.1	5	<.70
	2	1.2	2-3	11.6	5	<.05
	3	1.6	1-3	15.0	5	.01
4652^{Vg}	1	0.2	1-2	2.0	2	>.30
	2	0.0	2-3	.9	1	>.30
	3	0.1	1-3	1.1	2	<.30
4620^R	1	0.0	1-2	4.9	2	<.10
	2	0.4	2-3	6.7	5	>.20
	3	1.4	1-3	10.4	5	>.05

[a]F-all progeny fertile.

Chi square values for dates 1 and 3 approached significance in the cross involving the Reid source of male-sterility, but there appeared to be no real effect brought about through time of planting.

Only dates 1 and 3 of A158^{S} x Tx127C differ significantly in degree of expression of male-sterility, considering all crosses in Table 11. P values for the cross with J.C.33-16 x Mo2RF are quite high, as will be noted in more of the tables immediately following, indicating the lack of any important effect resulting from time of planting.

Table 12 indicates that expression of male-sterility in crosses between some cytoplasmic sources and TxGJ39 is markedly influenced by the changes in environment associated with date of planting. Apparently the partial fertility restoring contribution from TxGJ39 is more highly influenced by environmental factors encountered through different planting dates than are the fertility restoring genes of other testers.

Crosses of WF9^{S}, A158^{S}, and 4652^{Vg} with TxGJ39 shed some pollen in the third date of planting. This was in direct contrast to the situation for the first two dates. Dates 2 and 3, and 1 and 3 are different in the three-way cross (M1984 x M14) x TxGJ39. All dates differ in the cross involving 4620^{R} where complete fertility is restored in the last planting, but some sterile plants and partially fertile plants occur in the first two dates of planting.

The effect of K64 expressed in the F_1 cross seems to be quite variable over the three planting dates. This is particularly noticeable in Table 13 in the crosses with the Texas source where at each date partially fertile plants occur. Dates 1 and 3 are different in the cross with Tx203^{T} and the comparison of dates 2 and 3 approaches significance. Tx61M^{R} x K64

Table 11. Chi square values for tests of independence between date of planting effects on crosses involving five sources of cytoplasmic male-sterility with Tx127C, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S		--	--	--	--	--
$WF9^S$		--	--	--	--	--
$A158^S$	1	3.0	1-2	2.4	3	<.50
	2	2.5	2-3	5.0	3	<.20
	3	2.0	1-3	11.6	3	<.01
$A158^B$		--	--	--	--	--
$A158^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
$Tx203^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
$Tx61M^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
J.C.33-16 x Mo2RF	1	5.0 S&F	1-2	0.1	1	<.80
	2	5.0 S&F	2-3	0.6	1	<.50
	3	5.0 S&F	1-3	1.1	1	<.30
M1984 x M14	1	4.7 S&F	1-2	2.1	4	.70
	2	4.2 S&F	2-3	5.4	4	>.20
	3	5.0 S&F	1-3	2.6	3	<.50
4652^{Vg}	1	1.5	1-2	6.8	4	<.20
	2	1.2	2-3	5.3	5	>.30
	3	2.0	1-3	5.6	5	>.30
4620^R		--	--	--	--	--

[a]S&F-sterile and fertile plants in the same progeny.

Table 12. Chi square values for tests of independence between date of planting effects on crosses involving seven sources of cytoplasmic male-sterility with TxGJ39, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S	1	5.0	1-2	0.0		
	2	5.0	2-3	0.0		
	3	5.0	1-3	0.0		
$WF9^S$	1	5.0	1-2	0.0		
	2	5.0	2-3	42.0	1	<.01
	3	5.0PF	1-3	35.0	1	<.01
$A158^S$	1	5.0	1-2	0.0		
	2	5.0	2-3	38.0	1	<.01
	3	5.0PF	1-3	38.0	1	<.01
$A158^B$	1	4.8(one date)	--	--	--	--
$A158^T$		--	--	--	--	--
$Tx203^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
$Tx61M^T$	1	5.0F	1-2	0.0		
	2	5.0F	2-3	0.0		
	3	5.0F	1-3	0.0		
J.C.33-16 x Mo2RF	1	5.0S&F	1-2	0.0	1	>.80
	2	5.0S&F	2-3	0.1	1	>.70
	3	5.0S&F	1-3	0.0	1	>.80
M1984 x M14	1	4.9S,PF,&F	1-2	4.3	3	>.20
	2	4.8S,PF,&F	2-3	6.7	2	<.05
	3	5.0S&F	1-3	6.5	2	<.05
4652^{Vg}	1	5.0	1-2	0.0		
	2	5.0	2-3	39.0	1	<.01
	3	5.0PF	1-3	34.0	1	<.01
4620^R	1	5.0S,PF,&F	1-2	6.5	2	<.05
	2	5.0S,PF,&F	2-3	9.0	2	>.01
	3	5.0F	1-3	10.8	2	<.01

[a]S&F-sterile and fertile plants in the same progeny; S,PF,&F-sterile, partially fertile, and fertile plants in the same progeny.

Table 13. Chi square values for tests of independence between date of planting effects on crosses involving six sources of cytoplasmic male-sterility with K64, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S		--	--	--	--	--
$WF9^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^S$	1	0.5	1-2	5.7	5	>.30
	2	1.8 S&PF	2-3	8.3	5	<.20
	3	3.7 S&PF	1-3	15.0	4	<.01
$A158^B$		--	--	--	--	--
$A158^T$	1	1.6 PF	1-2	5.9	4	.20
	2	1.9 S&PF	2-3	2.1	4	.70
	3	1.7 S&PF	1-3	5.7	3	>.10
$Tx203^T$	1	3.5 PF	1-2	8.2	4	<.10
	2	2.6 S&PF	2-3	10.5	5	>.05
	3	2.0 S&PF	1-3	17.4	5	<.01
$Tx61M^T$	1	0.9 S&PF	1-2	13.7	3	<.01
	2	0.4 S&PF	2-3	13.5	4	<.01
	3	1.3 S&PF	1-3	5.7	4	>.20
J.C.33-16 x Mo2RF	1	3.8 S&F	1-2	8.3	5	.15
	2	4.2 S&F	2-3	6.4	2	<.05
	3	5.0 S&F	1-3	9.7	5	<.10
M1984 x M14	1	1.7	1-2	1.4	5	>.90
	2	1.6	2-3	9.4	5	<.10
	3	2.4	1-3	6.7	5	>.20
4652^{Vg}	1	0.0	1-2	0.0		
	2	0.0	2-3	3.8	1	.05
	3	0.2	1-3	3.6	1	>.05
4620^R	1	0.0	1-2	0.0		
	2	0.0	2-3	2.0	2	>.30
	3	0.4	1-3	2.3	2	>.30

[a]S&F-sterile and fertile plants in the same progeny; S&PF-sterile and partially fertile plants in the same progeny.

at date 1 is significantly different from the same F_1 at date 2. The same cross at dates 2 and 3 differs significantly.

In the 3-way cross involving J.C.33-16 x Mo2RF, lower P values are obtained in combination with K64 than with any of the other tester lines.

Only dates 1 and 3 are unlike in the F_1 cross of $A158^S$ x K64, but here again partially fertile plants occur in the progeny.

The last planting date of the Vg male-sterile source appears to possibly exsert more anthers than the first two dates but this variability is of doubtful significance.

There seems to be a greater change in expression of male-sterility from date 1 to date 2 than from 2 to 3 in Table 14 for crosses of A^S, $A158^S$, $A158^B$, and 4652^{Vg} with K55.

Dates 1 and 3 are markedly different in the 3-way cross (M1984 x M14) x K55. Table 15 lists significant chi square values for dates 1 and 2 and 1 and 3 of the $A158^S$ x M14 cross and for dates 1 and 3 of $A158^B$ x M14.

The early plantings of the Texas source crossed with M14 have lower anther exsertion mean scores than the later dates. The second and third and first and third dates of planting are different for $Tx203^T$ but only the first and third differ for $Tx61M^T$ with M14 as the male parent.

Each of the two lines converted to the U.S.D.A. source (Table 16), when crossed with Mo2RF, vary in degree of expression of male-sterility from date 1 to date 3.

All three dates are significantly different in the case of the 3-way cross (M1984 x M14) x Mo2RF. The incomplete male-sterility found in the M1984 x M14 single cross seems to be highly influenced by environmental conditions in several of the crosses with fertility differential testers.

Table 14. Chi square values for tests of independence between date of planting effects on crosses involving seven sources of cytoplasmic male-sterility with K55, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S	1	0.1	1-2	12.1	3	<.01
	2	1.2	2-3	5.2	3	>.10
	3	1.1	1-3	9.3	3	>.02
$WF9^S$	1	2.9	1-2	3.6	2	<.20
	2	3.2	2-3	1.4	2	<.50
	3	3.4	1-3	4.7	2	<.10
$A158^S$	1	0.0	1-2	23.9	3	<.01
	2	1.7	2-3	3.9	4	<.50
	3	2.1	1-3	38.2	4	<.01
$A158^B$	1	0.8	1-2	14.4	5	>.01
	2	2.6	2-3	5.8	5	>.30
	3	3.3	1-3	21.0	4	<.01
$A158^T$	1	5.0 F	1-2	0.0		
	2	5.0 F	2-3	0.0		
	3	5.0 F	1-3	0.0		
$Tx203^T$	1	5.0 F	1-2	0.0		
	2	5.0 F	2-3	0.0		
	3	5.0 F	1-3	0.0		
$Tx61M^T$	1	5.0 F	1-2	0.0		
	2	5.0 F	2-3	0.0		
	3	5.0 F	1-3	0.0		
J.C.33-16 x Mo2RF	1	5.0 S&F	1-2	0.3	1	.60
	2	5.0 S&F	2-3	2.5	1	>.10
	3	5.0 S&F	1-2	1.2	1	<.30
M1984 x M14	1	2.7	1-2	10.3	6	>.10
	2	4.6	2-3	7.0	4	>.10
	3	4.8	1-3	19.5	6	<.01
4652^{Vg}	1	0.8	1-2	15.2	3	<.01
	2	1.2	2-3	7.2	5	.20
	3	1.8	1-3	13.6	5	<.02
4620^R	1	1.3	1-2	3.7	5	>.50
	2	1.8	2-3	6.6	5	.25
	3	2.2	1-3	6.7	5	>.20

[a]S&F-sterile and fertile plants in the same progeny.

Table 15. Chi square values for tests of independence between date of planting effects on crosses involving six sources of cytoplasmic male-sterility with M14, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S	1	0.0	1-2	0.0		
	2	0.0	2-3	4.4	3	>.20
	3	0.3	1-3	3.2	3	>.30
$WF9^S$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$A158^S$	1	0.3	1-2	14.4	2	<.01
	2	1.1	2-3	4.2	3	>.20
	3	1.4	1-3	25.5	3	<.01
$A158^B$	1	0.4	1-2	1.7	3	<.70
	2	0.6	2-3	4.9	3	<.20
	3	1.3	1-3	10.7	3	>.01
$A158^T$		--	--	--	--	--
$Tx203^T$	1	2.7 PF	1-2	4.4	5	<.50
	2	2.5 PF	2-3	12.7	5	>.02
	3	1.0 PF	1-3	14.8	5	>.01
$Tx61M^T$	1	4.3 PF	1-2	6.5	4	<.20
	2	3.1 PF	2-3	5.4	5	>.30
	3	2.1	1-3	16.4	5	<.01
J.C.33-16 x Mo2RF	1	3.6 S&F	1-2	1.3	6	>.95
	2	3.8 S&F	2-3	3.8	6	.70
	3	4.2 S&F	1-3	6.7	6	>.30
4652^{Vg}	1	0.0	1-2	1.2	1	<.30
	2	0.2	2-3	4.4	2	>.10
	3	0.2	1-3	4.3	1	<.05
4620^R	1	0.1	1-2	0.5	1	<.50
	2	0.1	2-3	4.9	3	<.20
	3	0.7	1-3	5.7	3	>.10

[a]S&F-sterile and fertile plants in the same progeny.

Table 16. Chi square values for tests of independence between date of planting effects on crosses involving six sources of cytoplasmic male-sterility with Mo2RF, and anther exsertion mean score for each date

Source of sterility	Date	Mean score[a]	Planting dates compared	X^2	df	P
A^S		--	--	--	--	--
$WF9^S$	1	0.6	1-2	6.9	3	<.10
	2	1.4	2-3	1.7	3	<.70
	3	1.2	1-3	7.6	2	>.02
$A158^S$	1	0.4	1-2	3.0	2	>.20
	2	0.7	2-3	1.1	2	<.30
	3	0.8	1-3	7.4	2	>.02
$A158^B$	1	0.8	1-2	0.3	3	>.95
	2	0.9	2-3	9.7	5	<.10
	3	1.5	1-3	9.7	5	<.10
$A158^T$		--	--	--	--	--
$Tx203^T$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
$Tx61M^T$	1	0.0	1-2	0.0		
	2	0.0	2-3	0.0		
	3	0.0	1-3	0.0		
M1984 x M14	1	3.9 S&F	1-2	12.9	5	>.02
	2	3.2 S&F	2-3	8.1	2	<.02
	3	5.0 S&F	1-3	21.3	5	<.01
4652^{Vg}	1	0.3	1-2	1.5	3	<.70
	2	0.2	2-3	10.5	4	<.05
	3	1.1	1-3	10.1	4	<.05
4620^R	1	1.1	1-2	5.9	4	.20
	2	0.8	2-3	14.8	4	<.01
	3	2.7	1-3	14.4	4	<.01

[a]S&F-sterile and fertile plants in the same progeny.

When the Vg and the Reid sources are each crossed to Mo2RF, dates 2 and 3 and 1 and 3 are not the same.

One source of cytoplasmic male-sterility, that of Vg, did show environmental effect between dates 1 and 3 when crossed with inbred line Kys. These data are not shown in this series of tables. Most of the plants in this progeny were entirely sterile at all three dates, but a few plants in the third planting exserted anthers sufficient to justify a classification from 1 to as high as 4. The male-sterile source, 4652^{Vg}, is not a homozygous stock. Modifiers contributed by the female parent and segregating in the progeny of the cross with Kys present a possible explanation.

Relic Heterozygosity

As has been outlined in the section on materials and methods, individual plants of the lines Hy and C.I.7 were crossed to available plants of male-sterile lines and male-sterile single crosses. A number of different lines which had been converted to cytoplasmic male-sterility were used because of the small number of plants available of any one line. The single crosses were used only because of the shortage of male-sterile inbred line plants.

The individual Hy plants which were used to pollinate $A158^{S}$ tester plants are listed in Table 17. Mean scores for per cent of tassel area showing anther exsertion in the single crosses are given. Only ten Hy plants were outcrossed to the U.S.D.A. type of male-sterility. In each case all progeny were sterile but some anthers were visible, as shown by

the mean anther exsertion scores in Table 17. Not one of the ten plants tested had fertility restoring genes for the U.S.D.A. form of cytoplasmic male-sterility. The variation in mean anther exsertion scores can well be attributed to environmental influence or to scoring error.

Table 17. Degree of male-sterility expressed in the crosses between individual Hy plants and A158^{S}

Source of Hy seed lot	Male-sterile parent	Male parent	Mean anther exsertion score[a]	Degree of male sterility[b]
U.S.D.A. Exp. Sta., Beltsville	A158^{S}	x Hy 1-9	2	S
	"	x Hy 1-11	1	S
Illinois Agr. Exp. Sta.	"	x Hy 2-10	2	S
	"	x Hy 2-12	2	S
Kansas Agr. Exp. Sta.	"	x Hy 3-10	1	S
	"	x Hy 3-14	1	S
	"	x Hy 3-20	1	S
Missouri Agr. Exp. Sta.	"	x Hy 5-6	2	S
	"	x Hy 5-8	1	S
	"	x Hy 5-20	2	S

[a]1-trace anthers exserted; 2-anthers exserted on 25 per cent of tassel area.

[b]S-sterile.

Data from test crosses of individual Hy plants with the Texas source of cytoplasmic male-sterility are shown in Tables 18 and 19.

None of the 46 plants of inbred line Hy listed in Table 18 restored fertility in the F_1 generation when crossed to any of the lines which had been converted to the Texas form of male-sterility. Source of the Hy seed lots had no effect on degree of expression of male-sterility.

Table 18. Degree of male-sterility expressed in the crosses between individual Hy plants and inbred lines converted to the Texas type of cytoplasmic male-sterility

Source of Hy seed lot	Male-sterile parent	Male parent	Mean anther exsertion score[a]	Degree of male sterility[b]
U.S.D.A. Exp. Sta., Beltsville	A158^{T}	x Hy 1-9	0	S
	"	x Hy 1-11	0	S
	Os420^{T}	x Hy 1-2	0	S
	"	x Hy 1-13	0	S
	"	x Hy 1-16	1	S
Illinois Agr. Exp. Sta.	A158^{T}	x Hy 2-6	0	S
	"	x Hy 2-10	0	S
	"	x Hy 2-11	0	S
	"	x Hy 2-12	0	S
	Os420^{T}	x Hy 2-1	0	S
	"	x Hy 2-3	0	S
	"	x Hy 2-7	0	S
Kansas Agr. Exp. Sta.	A158^{T}	x Hy 3-10	0	S
	C106^{T}	X Hy 3-1	0	S
	"	x Hy 3-5	0	S
	"	x Hy 3-7	0	S
	"	x Hy 3-12	0	S
	"	x Hy 3-18	0	S
	I205^{T}	x Hy 3-26	0	S
	Os420^{T}	x Hy 3-2	1	S
Kentucky Agr. Exp. Sta.	C106^{T}	x Hy 4-3	0	S
	"	x Hy 4-4	0	S
	"	x Hy 4-6	0	S
	"	x Hy 4-7	1	S
	"	x Hy 4-8	0	S
	Os420^{T}	x Hy 4-28	0	S
	"	x Hy 4-40	0	S
	A158^{T}	x Hy 4-5	0	S
	I205^{T}	x Hy 4-30	0	S
Missouri Agr. Exp. Sta.	A158^{T}	x Hy 5-6	0	S
	"	x Hy 5-8	0	S
	"	x Hy 5-20	0	S
	L289^{T}	x Hy 5-2	0	S
	"	x Hy 5-10	0	S
	"	x Hy 5-11	0	S
	"	x Hy 5-12	0	S
	"	x Hy 5-17	0	S
	"	x Hy 5-19	0	S

[a]0-no anthers exserted; 1-trace anthers exserted.

[b]S-sterile.

Table 18. (Continued)

Source of Hy seed lot	Male-sterile parent	Male parent	Mean anther exsertion score	Degree of male sterility
Missouri Agr. Exp. Sta.	L289^{T}	x Hy 5-26	0	S
	"	x Hy 5-27	0	S
	"	x Hy 5-30	0	S
	"	x Hy 5-33	0	S
	"	x Hy 5-36	0	S
	"	x Hy 5-40	0	S
	"	x Hy 5-43	0	S
	"	x Hy 5-44	0	S

Table 19. Degree of male-sterility expressed in 3-way crosses between individual Hy plants and Texas cytoplasmic male-sterile single crosses

Source of Hy seed lots	Male-sterile parent	Male parent	Mean anther exsertion score[a]	Degree of male sterility[b]
U.S.D.A. Exp. Sta., Beltsville	(Tx203^T x 187-2)	x Hy 1-2	0.0	S
	"	x Hy 1-3	0.0	S
	"	x Hy 1-7	0.0	S
	"	x Hy 1-13	0.0	S
	"	x Hy 1-15	0.0	S
	"	x Hy 1-16	0.0	S
	"	x Hy 1-17	0.0	S
	"	x Hy 1-21	0.0	S
	(Tx203^T x Os420)	x Hy 1-14	0.0	S
Illinois Agr. Exp. Sta.	(Tx203^T x 187-2)	x Hy 2-1	0.0	S
	"	x Hy 2-2	0.0	S
	"	x Hy 2-3	0.0	S
	"	x Hy 2-4	0.0	S
	"	x Hy 2-13	0.0	S
Kansas Agr. Exp. Sta.	(Tx203^T x Os420)	x Hy 3-1	0.0	S
	"	x Hy 3-6	0.8	S,2PF
	"	x Hy 3-7	0.1	S,1PF
	"	x Hy 3-8	0.8	S,2PF
	"	x Hy 3-11	1.2	S,5PF
	"	x Hy 3-12	0.1	S,3PF
	"	x Hy 3-13	0.4	S,2PF
	"	x Hy 3-15	2.0	S,6PF
	"	x Hy 3-16	1.0	S,2PF
	"	x Hy 3-17	0.8	S,2PF
	"	x Hy 3-24	0.4	S,2PF
	"	x Hy 3-25	0.3	S,1PF
	"	x Hy 3-36	0.9	S,4PF
	"	x Hy 3-41	0.6	S,1PF
	(Tx173D^T x B10)	x Hy 3-3	3.5	S,12F
	"	x Hy 3-4	3.1	S,10F
	"	x Hy 3-5	3.9	S,14F
	"	x Hy 3-30	3.2	S,11F
	(Tx203^T x B14)	x Hy 3-40	1.0	S
Kentucky Agr. Exp. Sta.	(Tx203^T x B14)	x Hy 4-3	0.0	S
	"	x Hy 4-4	0.0	S
	"	x Hy 4-8	0.0	S
	"	x Hy 4-13	1.0	S

[a]0-no anthers exserted; 1-trace anthers exserted; 2-anthers exserted on 25 per cent of tassel area; 3-anthers exserted on 50 per cent of tassel area; 4-anthers exserted on 75 per cent of tassel area; 5-anthers exserted over entire tassel.

[b]S-sterile; S,2PF-sterile, except for 2 plants partially fertile; S,1PF-sterile, except for 1 plant partially fertile, etc.; S,12F-sterile except for 12 fertile plants, etc.

Table 19. (Continued)

Source of Hy seed lots	Male-sterile parent	Male parent	Mean anther exsertion score	Degree of male sterility
Kentucky Agr.Exp. Sta.	(Tx203^{T} x B14)	x Hy 4-14	1.0	S
	"	x Hy 4-25	0.0	S
	"	x Hy 4-29	0.0	S
	"	x Hy 4-41	0.0	S
	"	x Hy 4-44	0.0	S
	(Tx203^{T} x Oh45)	x Hy 4-16	0.0	S
Missouri Agr.Exp.Sta.	(Tx173D^{T} x Oh45)	x Hy 5-2	0.2	S
	"	x Hy 5-3	0.0	S
	"	x Hy 5-10	0.4	S,3PF
	"	x Hy 5-11	0.7	S,6PF
	"	x Hy 5-12	0.2	S,2PF
	"	x Hy 5-13	0.8	S,7PF
	"	x Hy 5-15	0.2	S,2PF
	"	x Hy 5-17	0.2	S,3PF
	"	x Hy 5-19	1.1	S,9PF
	"	x Hy 5-22	0.6	S,5PF
	"	x Hy 5-23	0.7	S,6PF
	"	x Hy 5-34	0.5	S,4PF
	"	x Hy 5-35	1.0	S,8PF
	(Tx203^{T} x Os420)	x Hy 5-4	1.0	S

Somewhat different results are obtained through use of a single cross as the male-sterile parent in test crosses (Table 19) with individual Hy plants as male parents. A progeny from a 3-way cross represents a segregating generation and in a number of progenies partially fertile plants occur. When single crosses Tx203^{T} x 187-2 and Tx203^{T} x B14 are used as female parents, only sterile plants are recovered in the crosses with all Hy plants tested. A different situation is encountered when the single cross Tx203^{T} x Os420 is used as the male-sterile tester. A fairly low anther exsertion score indicates essentially sterile plants, but in almost every cross involving Hy a few partially fertile plants are found in the progeny. The data recorded for the crosses of Hy plants with the male-sterile parent Tx173D^{T} x Oh45 are very similar to that involving Tx203^{T} x Os420. A few of the Hy plants were outcrossed to a male-sterile line and also to a male-sterile single cross. Progenies from the crosses involving Hy 3-7, Hy 3-12, Hy 5-10, Hy 5-11, Hy 5-12, Hy 5-17, and Hy 5-19 with the male-sterile lines consisted of only sterile plants, but progenies from the same Hy plants crossed with male-sterile single crosses included a few partially fertile plants in each instance. These data suggest that the Hy plants do not carry a major gene or genes for fertility restoration. Some of the lines making up the single crosses apparently possess minor genes which can affect the expression of male-sterility when in certain combinations with genes from Hy. When they segregate, as seems to occur in some of the 3-way crosses, partially fertile plants may be obtained in the progenies.

The single cross Tx173D^{T} x B10 is unusual in that it is entirely sterile, with no exsertion of anthers, yet when used as the female parent

in a 3-way cross with four different Hy plants produces completely fertile and completely sterile progeny. The Hy plants were selected at random from a population which apparently will not restore fertility to the Texas type of male-sterility. In fact, one of the plants, Hy 3-5, was also outcrossed to a C106^{T} plant and the progeny was entirely sterile. B10 must carry a factor, or factors, which when complemented by the genotype, of Hy, results in male-fertility. Evidently Tx173D is not responsible for this effect on sterility. When Tx173D^{T} x Oh45 is pollinated by Hy 5 plants (assuming Hy 5 plants are the same genotype as Hy 3 plants) no fertile plants are found in the progeny.

The individual C.I.7 plants which were tested for relic heterozygosity of fertility restoring genes are listed in Tables 20 and 21. The same procedure was followed as has been given in detail concerning the testing of Hy plants. Fewer tester plants of male-sterile lines and male-sterile single crosses were available, however, for crossing with C.I.7. Each of 13 C.I.7 plants tested on the U.S.D.A. form of male-sterility restored fertility, indicating that those C.I.7 plants were homozygous at the locus or loci which served to restore fertility to that source of male-sterility. Table 20 lists the individual plants tested on the Texas cytoplasmic male-sterile inbred lines.

According to the data listed in Table 20, only one of the 34 C.I.7 plants tested shows any indication of fertility restoration. The mean anther exsertion score and degree of male-sterility for the test cross of C.I.7 5-1 is very similar to that which could be expected with a male-sterile single cross tester. It is possible that a mistake had been made in recording the cross on the pollinating bag in the field, or that tags

Table 20. Degree of male-sterility expressed in the crosses between individual C.I.7 plants and inbred lines converted to the Texas type of cytoplasmic male-sterility

Source of C.I.7 seed lot	Male-sterile parent	Male parent	Mean anther exsertion score[a]	Degree of male sterility[b]
U.S.D.A. Exp. Sta., Beltsville	I205^T	x C.I.7 1-2	0.0	S
	"	x C.I.7 1-5	0.0	S
	"	x C.I.7 1-6	0.0	S
	Tx61M^T	x C.I.7 1-1	0.0	S
	"	x C.I.7 1-10	0.0	S
	"	x C.I.7 1-14	0.0	S
	"	x C.I.7 1-16	0.0	S
	"	x C.I.7 1-18	0.0	S
	"	x C.I.7 1-19	0.0	S
	"	x C.I.7 1-20	0.0	S
Illinois Agr. Exp. Sta.	1205^T	x C.I.7 2-6	0.0	S
	"	x C.I.7 2-8	0.0	S
	"	x C.I.7 2-13	0.0	S
	Tx61M^T	x C.I.7 2-17	0.0	S
	"	x C.I.7 2-20	0.0	S
	"	x C.I.7 2-22	0.0	S
Kansas Agr. Exp. Sta.	Tx61M^T	x C.I.7 3-7	0.0	S
	"	x C.I.7 3-22	0.0	S
	"	x C.I.7 3-25	0.0	S
	"	x C.I.7 3-30	0.0	S
	C106^T	x C.I.7 3-9	0.0	S
	"	x C.I.7 3-10	0.0	S
	"	x C.I.7 3-11	0.0	S
	"	x C.I.7 3-13	0.0	S
	"	x C.I.7 3-16	0.0	S
Kentucky Agr. Exp. Sta.	Tx61M^T	x C.I.7 4-7	0.0	S
	"	x C.I.7 4-10	0.0	S
	"	x C.I.7 4-19	0.0	S

[a] 0 - no anthers exserted; 1 - trace anthers exserted; 2 - anthers exserted on 25 per cent of tassel area; 3 - anthers exserted on 50 per cent of tassel area; 4 - anthers exserted on 75 per cent of tassel area; 5 - anthers exserted over entire tassel.

[b] S - sterile; S,5PF - sterile except for 5 plants partially fertile.

Table 20. (Continued)

Source of C.I.7 seed lot	Male-sterile parent	Male parent	Mean anther exsertion score	Degree of male sterility
Missouri Agr. Exp. Sta.	Os420^{T}	x C.I.7 5-1	0.4	S,5PF
	"	x C.I.7 5-4	0.0	S
	"	x C.I.7 5-8	0.0	S
	Tx61M^{T}	x C.I.7 5-23	0.0	S
	"	x C.I.7 5-27	0.0	S
	"	x C.I.7 5-28	0.0	S

Table 21. Degree of male-sterility expressed in 3-way crosses between individual C.I.7 plants and Texas cytoplasmic male-sterile single crosses

Source of C.I.7 seed lot	Male-sterile parent	Male parent	Mean anther exsertion score[a]	Degree of male sterility[b]
U.S.D.A. Exp. Sta., Beltsville	(Tx203^T x B6)	x C.I.7 1-3	0.1	S
	"	x C.I.7 1-7	0.1	S
Illinois Agr. Exp. Sta.	(Tx203^T x 38-11)	x C.I.7 2-2	0.0	S
	"	x C.I.7 2-3	0.1	S,1PF
	"	x C.I.7 2-5	0.1	S
	"	x C.I.7 2-10	0.0	S
	"	x C.I.7 2-18	0.0	S
	(Tx203^T x B14)	x C.I.7 2-14	0.2	S,3PF
	(Tx203^T x B10)	x C.I.7 2-15	0.4	S,3PF
Kansas Agr. Exp. Sta.	(Tx203^T x B6)	x C.I.7 3-1	1.3	S,8PF
	"	x C.I.7 3-2	1.3	S,8PF
	"	x C.I.7 3-3	0.7	S,6PF
	"	x C.I.7 3-4	0.3	S,4PF
	"	x C.I.7 3-5	0.8	S,7PF
	"	x C.I.7 3-6	0.8	S,7PF
	"	x C.I.7 3-8	0.5	S,6PF
	"	x C.I.7 3-14	0.6	S,8PF
	"	x C.I.7 3-19	0.5	S,6PF
Kentucky Agr. Exp. Sta.	(Tx173D^T x B10)	x C.I.7 4-1	0.4	S,1PF
	"	x C.I.7 4-4	0.2	S,3PF
	"	x C.I.7 4-5	0.1	S
	"	x C.I.7 4-8	0.0	S
	"	x C.I.7 4-9	0.1	S,2PF
	"	x C.I.7 4-11	0.2	S,3PF
	"	x C.I.7 4-14	0.2	S,3PF
	"	x C.I.7 4-20	0.5	S,3PF
	"	x C.I.7 4-24	0.2	S,3PF

[a] 0 - no anthers exserted; 1 - trace anthers exserted; 2 - anthers exserted on 25 per cent of tassel area; 3 - anthers exserted on 50 per cent of tassel area; 4 - anthers exserted on 75 per cent of tassel area; 5 - anthers exserted over entire tassel.

[b] S - sterile; S,1PF - sterile, except for 1 partially fertile plant, etc.

Table 21. (Continued)

Source of C.I.7 seed lot	Male-sterile parent	Male parent	Mean anther exsertion score	Degree of male sterility
Missouri Agr. Exp. Sta.	(Tx203^{T} x 38-11)	x C.I.7 5-11	0.0	S
	"	x C.I.7 5-13	0.0	S
	"	x C.I.7 5-14	0.0	S
	"	x C.I.7 5-15	0.0	S
	"	x C.I.7 5-16	0.0	S
	"	x C.I.7 5-17	0.0	S
	(Tx173D^{T} x B10)	x C.I.7 5-9	0.0	S
	"	x C.I.7 5-20	0.0	S

became mixed at time of shelling. Since every other individual plant of C.I.7 and of Hy tested on a male-sterile inbred line produced male-sterile progeny, the entry for C.I.7 5-1 may be assumed to represent an error.

Apparently all C.I.7 plants tested were homozygous recessive at the locus or loci involved in restoration of fertility to the Texas type of cytoplasmic male-sterility, and were homozygous dominant (based on a sample of 13 plants) for the gene or genes which effect restoration of fertility to the U.S.D.A. form of cytoplasmic male-sterility.

There were no differences among the 5 C.I.7 seed lots in expression of male-sterility.

Use of a male-sterile single cross in place of a male-sterile inbred line as a tester for C.I.7 plants results in a slightly different array of data. Table 21 indicates presence of modifier genes in some of the lines making up the single cross female parents, as was the case in tests of Hy plants. C.I.7 plants tested bear no major genes capable of restoring fertility. Apparently certain combinations of the modifiers bring about partial fertility in a few plants in some of the 3-way crosses. These genes merely modify the degree of expression of male-sterility. The single cross $Tx203^{T}$ x 38-11 seems to effectively sterilize the C.I.7 plants tested. A very few partially fertile plants occur in the progeny of the 3-way crosses involving $Tx203^{T}$ x B14 and $Tx203^{T}$ x B10 but since only one C.I.7 plant was tested in each case, little can be said concerning modifier genes. When $Tx173D^{T}$ x B10 is used as the tester parent a few partially fertile plants occur in some of the progenies, but no fertile plants, as was the case when Hy plants were used as the pollen parents.

Further tests could possibly reveal a difference in the modifier background of C.I.7 plants from the U.S.D.A. station at Beltsville and C.I.7 plants from the Kansas Agricultural Experiment Station. When $Tx203^T$ x B6 is used as the tester on C.I.7-3 plants a few partially fertile plants are found in the progeny. When the same tester is used on C.I.7-1 plants there are no partially fertile progeny, and the mean anther exsertion score is noticeably lower. Unfortunately only two C.I.7-1 plants were tested, so no conclusions can be drawn, but further testing may be of interest. It is possible that the two "sub-lines" of C.I.7 do differ in genes which modify expression of male-sterility as well as in genes which affect such characteristics as maturity and top-firing. Indications are, however, that no appreciable heterozygosity persists within any one "sub-line".

Morphological Study of Anther and Pollen Development in M1984 x M14 As Contrasted to That in M14 x M1984

Comparative growth rates of glume length are similar in the incompletely male-sterile single cross M1984 x M14 and in the fertile reciprocal cross M14 x M1984. Figure 1 shows the growth curves for glume length from sampling date 1 at meiosis (pedicellate floret) to sampling date 5 at anthesis (pedicellate floret) for the two single crosses. The greatest increment in length of glumes, for the period sampled, is added between dates 1 and 2. Evidently, maximum glume length has been attained by date 2, four days after meiosis, and subsequent variability merely reflects sampling error. Table 22 presents the analysis of variance for the attribute glume length. The mean square for dates, tested against the mean

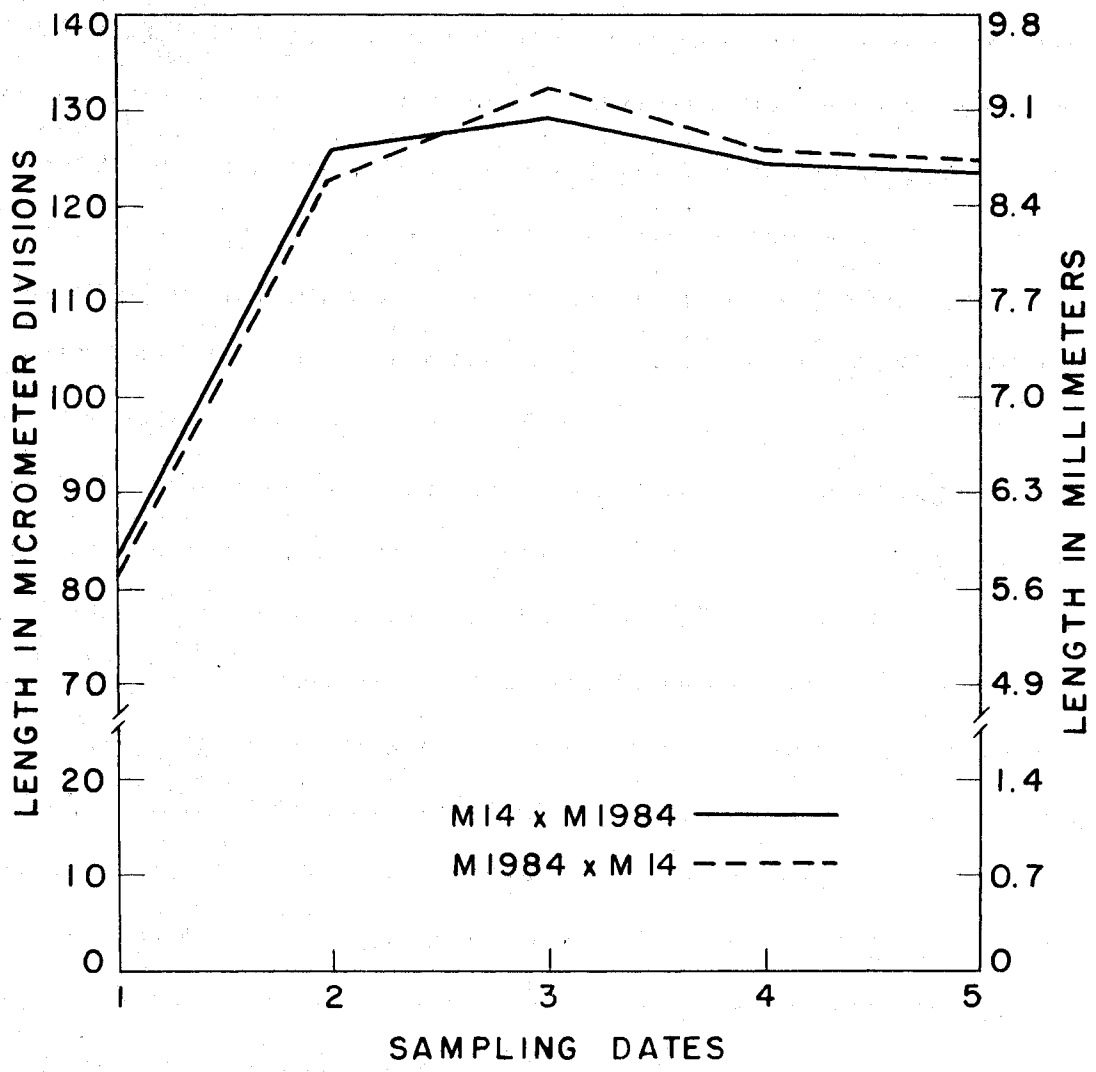

Figure 1. Glume length in the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates.

square for crosses within dates, is highly significant, indicating a true difference in length of glumes at different dates. Crosses within dates, tested against the mean square for plants within crosses, are not significantly different. The cytoplasmic-genotypic interaction responsible for the difference in degree of male-fertility between the two crosses has no important effect on glume length.

Table 22. Analysis of variance of glume length in the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates

Source of variation	Degrees of freedom	Mean squares for glume length[a]
Dates	4	45,788.12**
Crosses within dates	5	156.69
Crosses within date 1	1	20.00
" " " 2	1	238.00
" " " 3	1	350.21
" " " 4	1	91.88
" " " 5	1	83.33
Plants within crosses	110	197.32**
Spikelets within plants	480	38.31
Total	599	---

[a]** F exceeds the 1 per cent level of probability.

Figure 2 presents growth curves for anther length in both florets within a spikelet. The development of the sessile floret is appreciably retarded compared to the pedicellate floret of the same spikelet. The two curves representing the normal increase in length of anthers in the fertile cross are very nearly parallel, while the same comparison in the incompletely male-sterile cross shows that after date 3 there is a marked

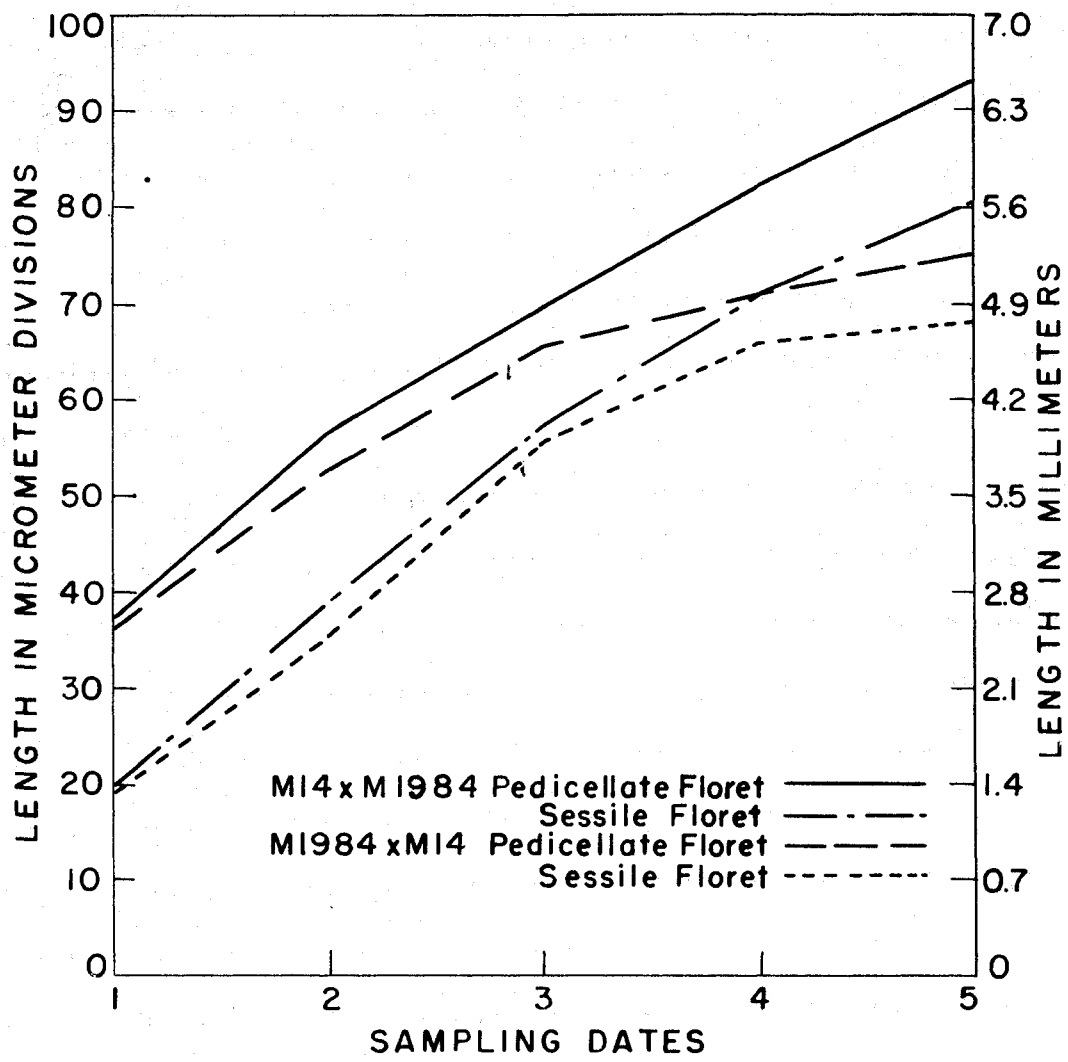

Figure 2. Anther length in the pedicellate and in the sessile florets for the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates.

decrease in rate of growth of the pedicellate floret, but the corresponding decrease in growth rate for the sessile floret does not come until the fourth date. This suggests that at some stage of development of the anthers a critical point is reached at which the cytoplasmic-genotypic interaction system expresses its greatest effect. In the incompletely male-sterile cross the marked change in growth rate occurs after the anthers have reached a length of approximately 65 micrometer divisions.

The anthers are slightly shorter in M1984 x M14 than in the normal reciprocal cross at date 1 for both florets. The two crosses differ significantly in anther length at date 2 (Table 23) for both florets. The difference between crosses is highly significant for length of anthers from the pedicellate florets for dates 3, 4, and 5. The growth curves in Figure 2 indicate a marked decrease in rate of growth in length of anthers for the incompletely male-sterile single cross after date 3. The fact that the mean square for crosses within date 3, in Table 23, for the sessile floret is not significant could possibly be attributed to sampling error since the difference between crosses is highly significant for both dates 4 and 5.

The growth curves for anther diameter in both florets are shown in Figure 3. The anthers in M1984 x M14 are of smaller diameter than those in the reciprocal cross at all five dates. Mean squares for crosses within each date for the pedicellate floret listed in Table 24 are highly significant. The rate of increase in anther diameter shows a sharp rise at date 3 for the normal single cross, whereas rate of growth in diameter of the anthers in the incompletely male-sterile single cross is quite

Table 23. Analysis of variance of anther length in the pedicellate and in the sessile florets for the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates

Source of variation	Degrees of freedom	Mean squares for pedicellate floret[a]	Mean squares for sessile floret[a]
Dates	4	128,897.09**	190,049.24**
Crosses within dates	5	8,868.59**	3,338.69**
Crosses within date 1	1	108.90	87.03
" " " 2	1	1,432.00*	986.71*
" " " 3	1	1,707.38**	273.88
" " " 4	1	11,537.34**	2,205.23**
" " " 5	1	29,557.34**	13,140.62**
Plants within crosses	110	211.75**	231.68**
Spikelets within plants	480	29.19**	34.86**
Anthers within spikelets	1200	7.09	3.99
Total	1799	---	---

[a]** F exceeds the 1 per cent level of probability.
* F exceeds the 5 per cent level of probability.

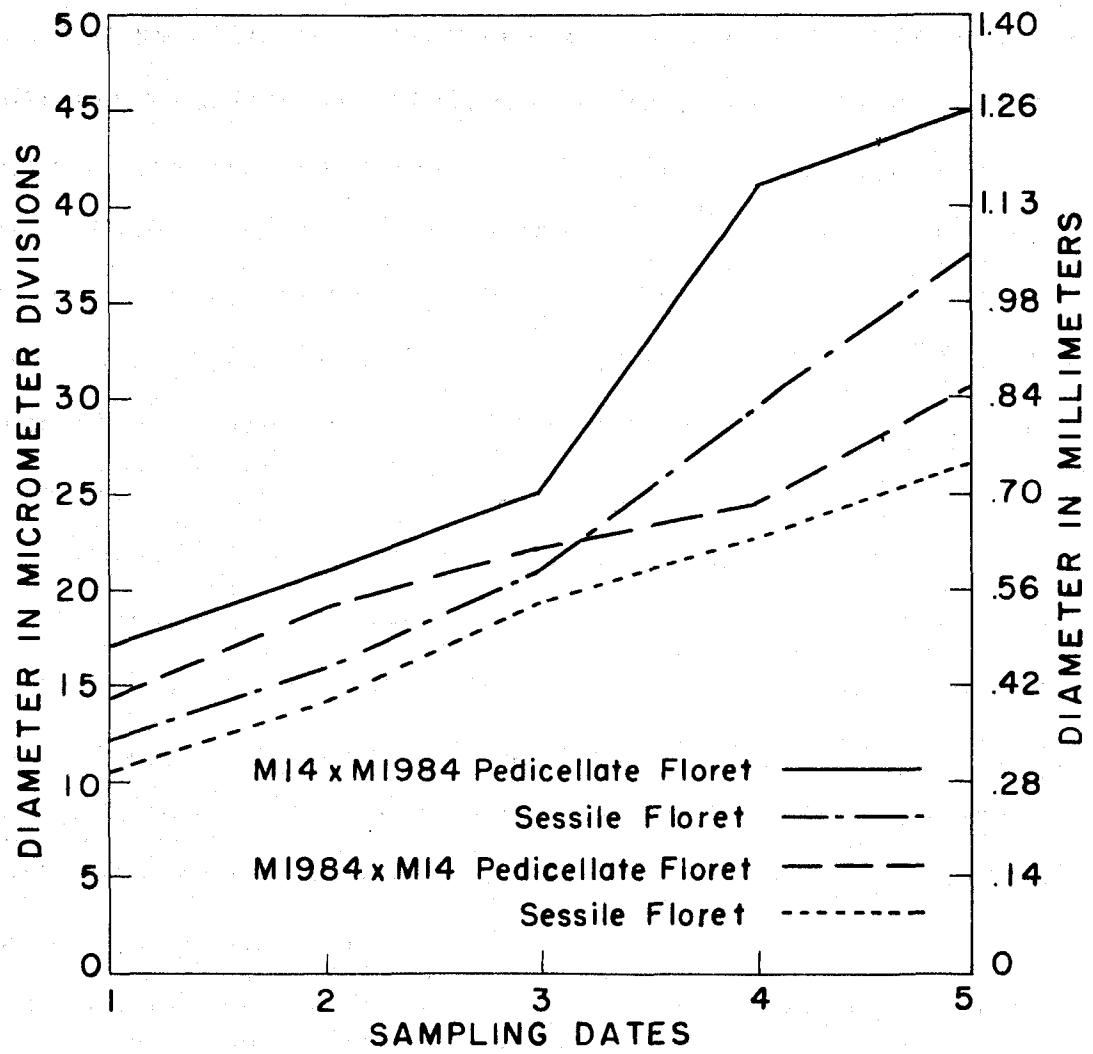

Figure 3. Anther diameter in the pedicellate and in the sessile florets for the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates.

Table 24. Analysis of variance of anther diameter in the pedicellate and in the sessile florets for the reciprocal single crosses M14 x M1984 and M1984 x M14 for five sampling dates

Source of variation	Degrees of freedom	Mean squares for pedicellate floret[a]	Mean squares for sessile floret[a]
Dates	4	30,317.37	25,419.34*
Crosses within dates	5	8,919.31**	3,256.64**
Crosses within date 1	1	711.21**	224.04*
Crosses within date 2	1	399.00**	372.10**
" " " 3	1	640.00**	222.47*
" " " 4	1	24,370.68**	5,010.14**
" " " 5	1	18,475.67**	10,454.44**
Plants within crosses	110	48.01**	42.04**
Spikelets within plants	480	10.55**	9.22**
Anthers within spikelets	1200	2.73	1.91
Total	1799	---	---

[a]** F exceeds the 1 per cent level of probability.
* F exceeds the 5 per cent level of probability.

consistent throughout the five sampling dates. Mean squares in Table 24 for the sessile floret indicate significant differences between crosses at dates 1 and 3 and highly significant differences at dates 2, 4, and 5.

The anthers of M1984 x M14 did not attain the same length nor the same diameter as those of the fertile cross M14 x M1984. Figure 4 shows comparative photographs taken of anthers from both crosses at each sampling date.

Microscopic examination of the pollen grains at the time pollen diameter measurements were taken revealed the presence of some grains in M1984 x M14 which did not stain in iodine solution. No staining reaction occurred in the material representing samples from either cross before date 3, eight days after meiosis. Table 25 lists the average degree of iodine stain observed in each of 12 plants of M1984 x M14 and of 12 plants of M14 x M1984. No pollen grains showed indication of starch deposition in M1984 x M14 at date 3. All plants of M14 x M1984 sampled at the same date contained pollen grains which stained to some degree, indicating that deposition of starch granules had begun. There was, however, considerable range in degree of staining observed from plant to plant. All pollen grains (disregarding a very small number which did not stain) from plants of the fertile cross were partially stained at this date. Plants of the incompletely male-sterile cross at date 4 contained some pollen grains which were entirely stained, or very nearly so, and some grains which took no stain at all. Practically all pollen of the fertile reciprocal cross stained in full at this stage of development. Observations from staining tests of pollen grains at date 5 indicated much the same as the data from date 4. Approximately one-half of the pollen grains of M1984

Figure 4. Photographs of anthers removed from tassel spikelets at each of five sampling dates. Incompletely male-sterile single cross M1984 x M14, left; fertile reciprocal cross M14 x M1984, right; first sampling date, top row; second sampling date, second row; third sampling date, third row; fourth sampling date, fourth row; and fifth sampling date, bottom row.

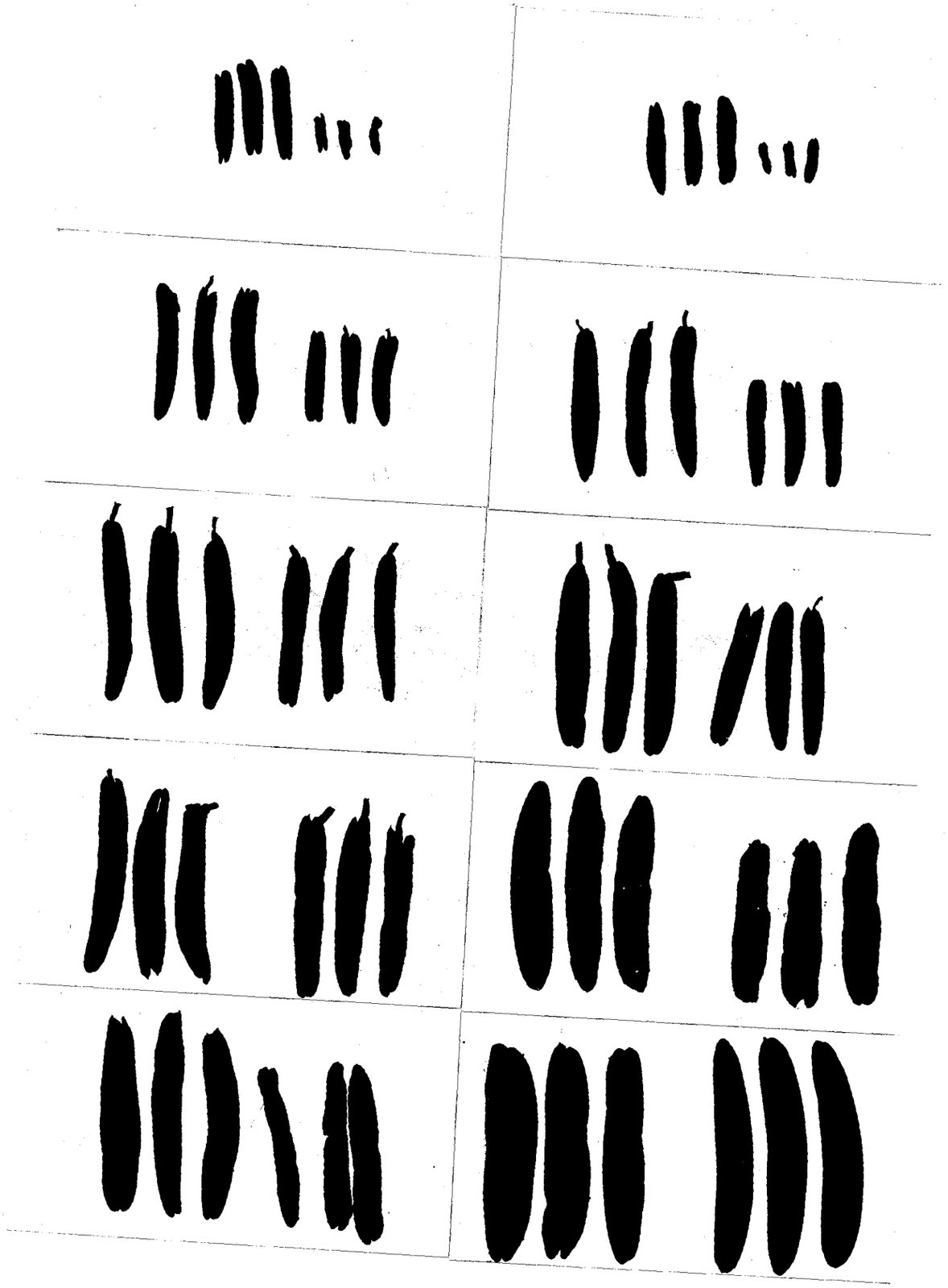

Table 25. Degree of iodine stain observed in the pollen grains from twelve plants of each of the reciprocal single crosses, M14 x M1984 and M1984 x M14.

	Date 3		Date 4		Date 5	
	M1984 x M14[a]	M14 x M1984[b]	M1984 x M14[c]	M14 x M1984[c]	M1984 x M14[c]	M14 x M1984[c]
Plant 1	---	30.0	38.2	99.0	43.2	99.0
2	---	14.0	51.6	98.2	50.0	99.0
3	---	34.0	43.2	99.0	50.0	99.0
4	---	71.0	32.6	99.0	46.6	99.0
5	---	58.0	60.0	98.2	50.0	99.0
6	---	15.0	50.0	98.2	43.2	99.0
7	---	63.0	33.2	99.0	46.6	99.0
8	---	6.0	51.6	97.4	50.0	99.0
9	---	8.0	50.0	98.2	50.0	99.0
10	---	7.0	50.0	99.0	50.0	99.0
11	---	5.0	50.0	99.0	50.0	99.0
12	---	79.0	37.6	99.0	46.6	99.0
Mean		32.7	45.7	98.6	48.0	99.0

[a]Pollen grains do not stain.

[b]All pollen grains partially stained, the amount of stain expressed in per cent.

[c]Per cent of pollen grains entirely stained.

x M14 were fully stained, and the remainder showed no stain. Virtually all pollen of M14 x M1984 stained completely. Photomicrographs of pollen samples taken from both crosses at each of the last three sampling dates are shown in Figure 5.

Table 26 lists the mean diameter of pollen grains from each of the reciprocal single crosses for the last three sampling dates. At date 3 only one size was noticeable in the M1984 x M14 pollen but by date 4 two sizes were present. The smaller grains did not stain nor did they increase in diameter after date 3, as indicated in Figure 6. Apparently the change brought about, which eventually became expressed in the form of incomplete male-sterility of the tassel, affected individual pollen grains of a specific genic complement at about the same time the development of the anthers was influenced. Those pollen grains in the same cross which did stain increased in size from date 3 through date 5 but did not seem to attain full size in comparison to those of the fertile cross, M14 x M1984. Mean squares for crosses within dates listed in Table 27 are highly significant at dates 3 and 5, but indicate no difference in pollen size between crosses at date 4. Apparently the normal pollen grains in M1984 x M14 were handicapped in the last stages of development, possibly a direct result of the failure of the anthers to have developed normally. Highly significant differences in pollen diameter observed at dates 4 and 5 in M1984 x M14 are indicated by the sizes in dates mean squares of Table 28.

Appendix Tables 33 to 42, inclusive, list individual plant measurements for glume length, mean anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14 and the fertile reciprocal cross M14 x M1984, at each of the five sampling dates.

Figure 5. Photomicrographs of pollen samples. Incompletely male-sterile single cross M1984 x M14, left; fertile reciprocal cross M14 x M1984, right; third sampling date, top row; fourth sampling date, middle row; fifth sampling date, bottom row.

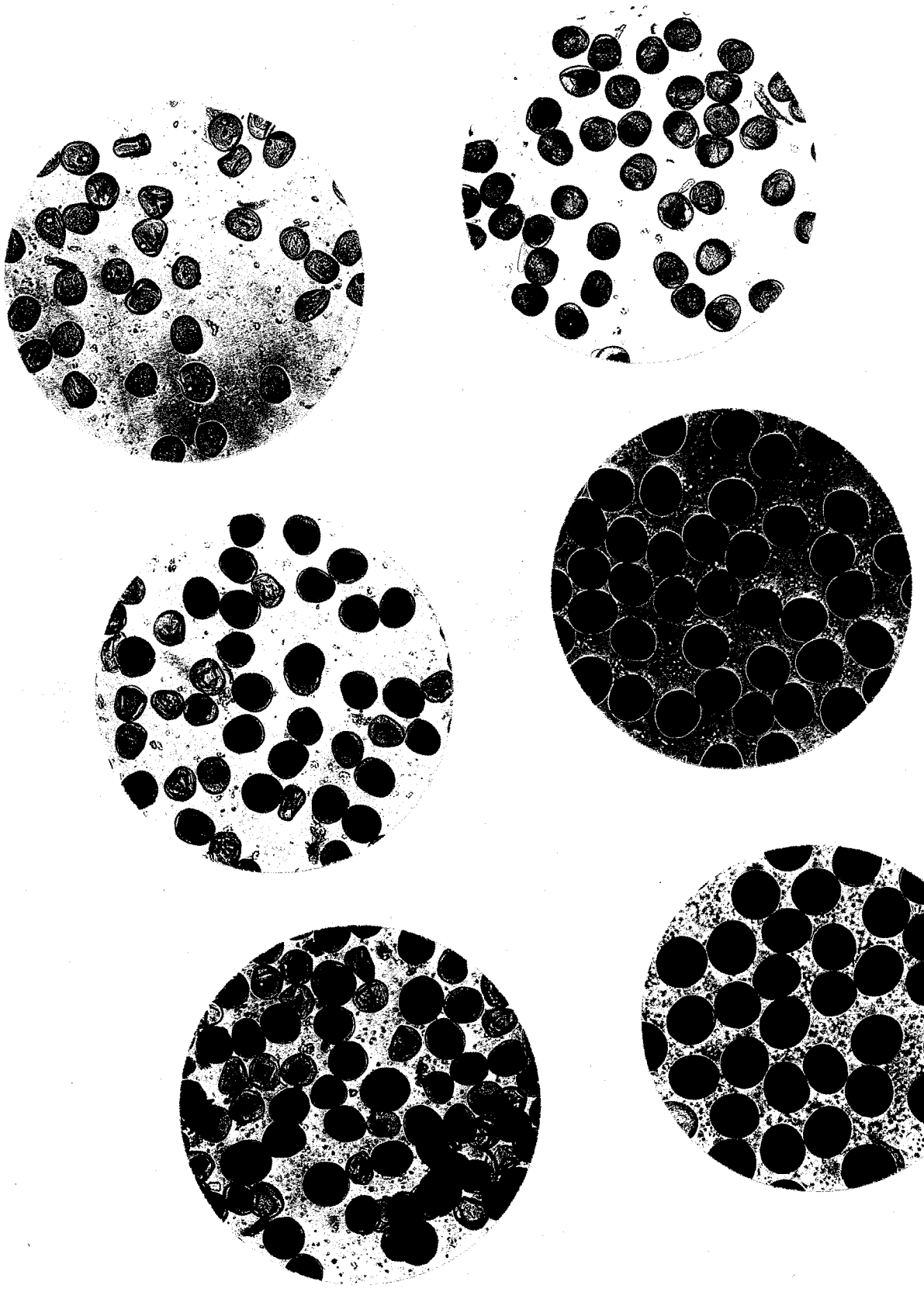

Table 26. Mean diameter of the pollen grains from twelve plants of each of the reciprocal single crosses, M14 x M1984 and M1984 x M14, expressed in micrometer divisions

	Date 3		Date 4			Date 5		
	M1984xM14	M14xM1984	M1984xM14[a]	M1984xM14	M14xM1984	M1984xM14[a]	M1984xM14	M14xM1984
Plant 1	9.1	10.0	9.6	10.4	11.4	9.4	11.8	13.3
2	9.6	9.8	9.5	11.0	10.5	9.1	12.0	12.9
3	8.9	10.0	9.5	10.7	10.9	9.2	11.0	13.1
4	9.3	10.0	9.5	10.2	11.1	9.0	11.5	13.6
5	9.0	10.0	9.6	11.0	11.1	9.5	11.5	12.8
6	9.4	9.9	9.5	10.9	10.7	9.4	11.0	13.5
7	9.9	10.0	9.5	11.0	11.3	9.1	10.8	13.7
8	9.7	9.6	9.5	11.0	10.3	9.0	11.3	13.3
9	9.7	9.5	9.5	11.0	10.1	9.2	11.5	13.5
10	9.5	9.7	9.1	11.0	10.7	9.5	11.3	13.0
11	9.4	9.8	9.5	10.5	10.6	9.5	11.7	12.5
12	9.8	10.0	9.3	10.1	10.9	9.1	11.5	14.4
Mean	9.4	9.9	9.5	10.7	10.8	9.2	11.4	13.3

[a]Diameter of pollen grains which do not stain with iodine.

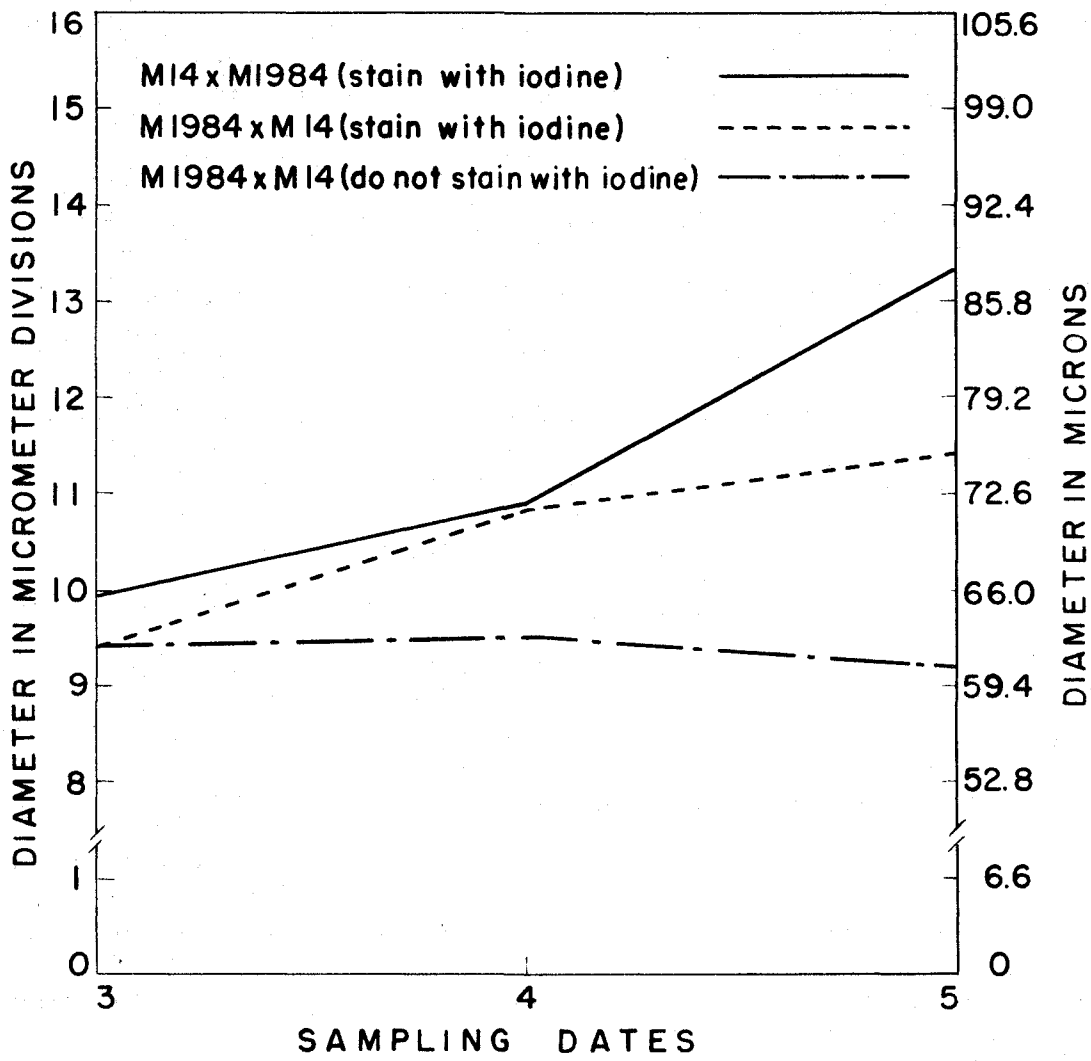

Figure 6. Pollen grain diameter in the reciprocal single crosses M14 x M1984 and M1984 x M14 for three sampling dates.

Table 27. Analysis of variance for diameter of normal pollen in the reciprocal single crosses M14 x M1984 and M1984 x M14 at three sampling dates

Source of variation	Degrees of freedom	Mean squares for pollen diameter[a]
Dates	2	221.60
Crosses within dates	3	37.56**
Crosses within date 4	1	5.21**
" " " 5	1	.13
" " " 6	1	107.35**
Plants within crosses	66	.65**
Spikelets within plants	288	.12
Total	359	

[a] ** F exceeds the 1 per cent level of probability.

Table 28. Analysis of variance of pollen diameter in the single cross M1984 x M14 for pollen grains which stain with iodine solution and those which do not stain, at two sampling dates

Source of variation	Degrees of freedom	Mean squares for pollen diameter[a]
Dates	1	3.15
Sizes within dates	2	93.95**
Sizes in date 4	1	48.13**
" " " 5	1	139.75**
Plants within sizes	44	.37**
Spikelets within plants	192	.09
Total	239	---

[a] ** F exceeds the 1 per cent level of probability.

A preliminary cytological investigation of stages in M1984 x M14 as early as pachytene and as late as formation of quartets of microspores failed to reveal any abnormalities in the meiotic process.

Genetic Analyses

Incomplete male-sterility of M1984 x M14

Data from preliminary genetic tests suggested the hypothesis that this type of male-sterility was the result of a cytoplasmic component from the line M1984 interacting with an incompletely dominant gene from M14 plus minor effects of a series of modifier genes contributed by both parents. Effects brought about by this cytoplasmic-genotypic interaction system were expressed through abnormal anther development in all florets of the tassel, and in the apparent inviability of approximately one-half of the pollen grains produced by F_1 plants.

The marked difference noted between the male-fertile single cross M14 x M1984 and the incompletely male-sterile reciprocal cross M1984 x M14 is an indication of a cytoplasmic influence upon this type of male-sterility. The F_1 generations were both grown for two years at Ames. The M14 x M1984 single cross was male-fertile in 1952 and except for a late planting was male-sterile in 1953. Anthers were exserted over the entire tassel but all were small and shriveled. F_2 generations were grown of each cross both years. The M14 x M1984 F_2 was fertile, just as was the F_1. The M1984 x M14 F_2 segregated for male sterility. Table 29 gives the classification of individual plants, based on tassel appearance, for the segregating generations grown in 1953.

Table 29. Number of plants within each male-sterile or male-fertile class in segregating generations of M1984 x M14, for designated planting dates

Class[a]	M1984 x M14 F_2		(M1984 x m14)xM14		(M1984 x M14)xM1984		M1984x(M1984 x M14)		M1984x(M14 x M1984)
	May 9	May 26	May 9	May 30	May 9	May 30	May 9	May 30	May 30
	Number of plants								
0	28	46	126	34	6	--	2	--	4
1	11	14	17	16	2	--	1	--	3
2	9	12	6	6	-	--	2	--	8
3	11	15	6	8	4	1	3	--	1
4	22	20	10	4	4	5	3	1	3
4pf	4	3	--	--	-	--	-	--	-
5	14	30	9	1	11	14	8	1	6
5pf	25	30	2	3	2	10	19	9	2
5f	63	28	7	1	7	13	30	19	5
Total	187	198	183	73	36	43	68	30	32

[a]0 - no anthers exserted, no pollen shed; 1 - trace anthers exserted, no pollen shed; 2 - 25 per cent anthers exserted, no (or trace) pollen shed; 3 - 50 per cent anthers exserted; no (or trace) pollen shed; 4 - 75 per cent anthers exserted, no (or trace) pollen shed; 4pf - 75 per cent anthers exserted, up to 25 per cent pollen shed; 5 - 100 per cent anthers exserted, no (or trace) pollen shed; 5pf - 100 per cent anthers exserted, up to 75 per cent pollen shed; 5f - 100 per cent anthers exserted, up to 100 per cent pollen shed.

Previous microscopic examination of pollen from M1984 x M14 plants had revealed two types: large pollen grains which stained with iodine, and smaller grains which did not stain and apparently were non-viable. The two kinds occurred in approximately equal frequency, suggesting the possibility of a single gene difference. Since male-sterile F_1 plants were produced only when M14 was used as the male parent, and all these F_1 plants were uniformly male-sterile, it appears that M14 contributed a dominant effect conditioning male-sterility, but only in the presence of M1984 cytoplasm. The genotype for M1984 might be designated ss, and the genotype for M14 as SS. The male sterile F_1 plants would then be Ss. In selfing an F_1 plant, if one assumes that only the staining pollen grains are functional and that they carry the s gene, the F_2 population would be expected to consist of fertiles and steriles in the proportion of 1:1. In considering the May 9 planting date of M1984 x 14 F_2, Table 29, if classes 4f, 5pf, and 5f are included as fertiles, the ratio is 92 fertile to 95 sterile plants. The May 26 planting of the F_2 reached the stage of anthesis during hot dry weather. This may in part explain the difference between the F_2 populations at the two planting dates. Chi square for a test of independence between classifications for the two dates was highly significant, with a P value of less than .01, indicating marked environmental influence on expression of male-sterility. At each planting date the extreme or near extreme classes had higher frequencies than the intermediate classes. This is suggestive of a major gene, along with a series of modifiers. The intermediates could well be due to the expression of modifiers segregating independently of the postulated S gene.

The presence of completely sterile types in the F_2 may be explained by assuming that an occasional S pollen grain produced by an F_1 plant may develop sufficiently to be viable. Sterile F_2 plants showing no anther extrusion may be of the genotype SS. Plants showing no anther extrusion may also be represented by the Ss genotype plus the effect of modifiers which shift expression toward an extreme type of sterility.

The data in Table 29 indicate a definite predominance of completely male-sterile plants in the backcross of the F_1 as the female parent to M14, suggesting the possibility of incomplete dominance of the S gene. The plants in the 0 class are presumed to be either the SS or Ss genotypes interacting with the series of modifiers. Although the expression of male-sterility in the two planting dates differed significantly according to a chi square test for independence, the trend toward sterility was predominant in each. When M14 was used as the pistillate parent in the same backcross all progeny were fertile, indicating that the cytoplasmic component from M1984 was necessary for expression of male-sterility.

In the backcross to M1984 as pollen parent, Table 29 indicates an even distribution in the four extreme classes for the first planting date, but a trend toward fertility in the second date. The second planting of the backcrosses to M1984 was later in maturity than most second planting F_2 plants and reached the stage of anthesis during a period in which temperatures ranged from approximately 70° to 80° F. The population size was small for both dates of planting of (M1984 x M14) x M1984 but one might postulate that the data, although somewhat masked by modifiers, simulate a 1:1 ratio expected in a backcross to a simple recessive. When

the single cross M1984 x M14 was used as the pollen parent in the backcross, the trend toward fertility was pronounced as would be expected if only the g pollen grains generally were viable. When the same backcross was made with the reciprocal single cross (M14 x M1984) as the male parent, the distribution among classes was more uniform, as would be expected if both types of pollen grains were functional. Chi square for a test of independence between the M1984 x (M1984 x M14) and the M1984 x (M14 x M1984) populations was highly significant with a P value of less than .01. Larger sized populations would have been desirable.

Inbred line M1984, when crossed with K63 as pollen parent, produced completely male-sterile progeny. No anthers were exserted and no pollen shed by the F_1 plants. Apparently a gene (or genes) carried by K63 had an even greater sterilizing effect on the F_1 than had the line M14. None of the F_1 plants was examined to determine the type of pollen produced. Neither the reciprocal cross K63 x M1984 nor any backcrosses were available for study.

Other lines used as pollen parents in crosses with M1984 produced fertile progeny. One line, Mo2RF, when used as the male parent produced a partially fertile F_1. All plants were classified as being 50-75 per cent fertile.

A number of 3-way crosses were made involving the single cross M1984 x M14 as the female parent and each in the series of tester differential lines as male parents. The progeny from (M1984 x M14) x K63 were all sterile, although some plants did exsert a few anthers. Very nearly the same results were obtained in the 3-way cross (M1984 x M14) x WF9 as shown in Table 30. Chi square in a test of independence for expression of male-

sterility in the two 3-way crosses was not significant, having a P value of greater than .50. Based upon this comparison, it may be that WF9 used as the pollen parent in a single cross with M1984 would produce a male-sterile F_1. Two sterile plants from the 3-way cross (M1984 x M14) x WF9 were pollinated by normal WF9, and all progeny from both plants were male-sterile. There were 76 plants in one progeny and 56 in the other. It seems highly possible that WF9 can be sterilized through use of M1984 as the pistillate parent.

Table 30. Number of plants within each male-sterile or male-fertile class for 3-way crosses involving M1984 x M14 with K63 and with WF9 as pollen parents

Class[a]	(M1984 x M14) x K63	(M1984 x M14) x WF9
	Number of plants	
0	77	167
1	2	12
2	4	9
3	6	9
4	2	4
4pf	--	---
5	3	2
5pf	--	1
5f	--	3
Total	94	207

[a]Classes are the same as those described at the end of Table 29.

Vg and Reid male-sterility

Further study is needed on the genetics of these two sources of male-sterility. Data from preliminary tests indicate some type of selective fertilization in the case of a plant heterozygous for the fertility restoring gene or genes. It may be that only pollen of a certain genotype functions, similar to the action postulated for the M1984 x M14 type of

male-sterility. A working hypothesis has been formulated, assuming that in addition to the necessary cytoplasmic component the male-sterile plant was of the genotype SS or Ss. The lines which act as fertility restorers were homozygous for an allelic gene which is dominant to the S sterility gene, and designated by S'. The fertile F_1 is expected to be SS' if the male-sterile parent was homozygous at the S locus, or could be sS' if the male-sterile parent had been heterozygous. The presence of modifier genes is suggested in segregating generations of crosses with some of the fertility restoring lines.

The Vg male-sterile plants used in the crosses with the six possible restorer lines were progeny from one of the male-sterile plants of the original Vg sy j/sy v_{16} stock pollinated by sy gl_{16}, to eliminate the Vg gene. All plants were completely sterile. This progeny will hereafter be referred to as 4632^{Vg}. Male-sterile plants from the original Vg sy j/sy v_{16} stock will be referred to as 4652^{Vg}.

The F_1 of 4632^{Vg} x multiple tester segregated in a ratio of approximately 1 fertile:1 male-sterile, which would suggest either that the multiple tester stock was segregating for the restorer gene or genes, or that the sterile parent was heterozygous for a gene or genes which when complemented by the male parent genotype resulted in sterility, even if the male parent was homozygous for the restorer gene. A fertile F_1 plant was selfed to obtain an F_2 generation, and was also used as the pollen parent in a backcross to the male-sterile parent. A sterile F_1 plant was backcrossed to the multiple tester parent. All F_2 plants were completely fertile, as were all plants in each of the two backcross populations. Some type of selective fertilization such as a male gametophyte factor

seems to be the explanation. Pollen examinations were not made for the F_1 plants nor for any plants of the segregating generations. If the same type of gene action is involved as has been postulated for the M1984 x M14 single cross, except that an allelic gene from the multiple tester stock is dominant to the gene for sterility in the female parent, one would expect to find pollen grains of two different types in the fertile F_1 plants. The large sized completely filled grains would represent S' gametes, carrying the gene which restores fertility to the Vg type of sterility, while the small non-staining grains would carry the S gene for sterility. If the S grains are all non-functional, the F_2 plants would be either S'S' or S'S. Assuming complete dominance of the S' gene, all F_2 plants would be fertile. Examination of pollen of the 4632^{Vg} parent may indicate whether or not it is homozygous for the S gene. No pollen would be expected to stain if the genotype was SS, but approximately one-half of the grains would stain if it was Ss. If this condition were found, the assumption of complete dominance of the S gene would be necessary to explain the complete male-sterility of all plants of the 4632^{Vg} parent. This would still satisfy the requirement that all F_2 plants be fertile. The F_2 plants could be of the genotypes SS', S'S', or sS', any one of them being fertile. An F_3 generation under these assumptions would likewise be fertile.

The progeny of the backcross to the 4632^{Vg} parent, if the female parent was SS, would be SS', and all plants would be expected to be fertile. If the female parent was Ss, the backcross progeny would consist of the two genotypes SS' and sS', again all would be expected to be fertile. The predicted progeny of the backcross to the multiple tester

parent, if the female parent were Ss, would be SS', S'S', or sS'. In either case all backcross plants would be expected to be fertile. Classification could be based on pollen samples. SS' plants would be expected to have approximately one-half staining grains and the remaining ones non-staining. Plants of the genotypes S'S' and sS' should have all normal pollen grains.

If upon microscopic examination of the pollen of an F_1 plant there appeared to be no abnormal grains, the possibility exists that the S' genotype pollen grains may effect some type of selective fertilization over the pollen bearing the S allele.

In an attempt to find some other suitable explanation, consideration of two loci involved does not satisfy the requirements, assuming all pollen is viable. If the 4632^{Vg} parent was of the genotype SSff or Ssff and the multiple tester parent was ssFF (F gene epistatic to S) we would expect a fertile F_1. If a plant of the genotype SsFf were selfed to obtain an F_2 progeny, some male-sterile plants would be expected. About three-sixteenths of the plants would be S-ff and theoretically would be sterile. In the backcross to 4632^{Vg} approximately one-half of the progeny would be expected to be sterile, again of the genotype S-ff. All progeny of the backcross to the multiple tester parent should be fertile under these assumptions. If the original male-sterile parent plant had been heterozygous at the S locus, two types of F_1 plants could occur. If the F_1 plant that was selfed was ssFf, then all F_2 plants would be expected to be fertile. However, male-sterile plants would be expected in the backcross to the male-sterile parent in a ratio of 1 sterile:3 fertile. If

Sf pollen was non-viable, examination of the pollen of F_1 plants could reveal whether one gene or two is involved. Likewise, examination of pollen from F_2 and backcross plants would give further evidence of one gene or two.

In the cross 4620^R x multiple tester an entirely fertile F_1 was obtained, indicating that the multiple tester parent plant was homozygous for the fertility restoring gene or genes. The F_2 population of 135 plants was fertile, with the exception of one plant which was partially fertile. In the backcross to the male-sterile parent most of the progeny again were fertile. Six out of a total of 70 plants shed no pollen. Again, an explanation of a pollen sample from the individual plants of each generation would have been most helpful. The F_2 and backcross data suggest that an explanation similar to that for 4632^{Vg} may be given in the case of 4620^R x multiple tester. The one partially fertile F_2 plant and the six sterile plants of the backcross to 4620^R may be the result of an occasional S pollen grain developing far enough so that it may effect fertilization. The genotype of these plants could then be SS or possibly SS' plus the effect of modifiers. It is difficult to explain the presence of seven out of 33 sterile plants in the backcross to the restorer parent. The multiple tester plant used as male in this backcross may have been heterozygous for the restorer gene. If so, the backcross may have been SS' x S's. Under this assumption one-fourth of the progeny could be expected to be male-sterile (Ss).

If microscopic examination reveals no difference between pollen grains from F_1, F_2, or backcross plants there is a possibility, as in the

4632^{Vg} study, that a male gametophyte factor is responsible for the preponderance of fertiles in the populations involving the Reid source of male-sterility.

It is not known whether the same locus is involved in the crosses of 4632^{Vg} and of 4620^{R} with the multiple tester stock, but on the basis of the F_2 and backcross data it can be considered a possibility. The difference between the segregating generations involving 4632^{Vg} and 4620^{R} may be in the background of modifiers.

The hypothesis of two loci being involved does not seem to be a suitable explanation for the Reid male-sterile x multiple tester data. The expectations as outlined in the section on the Vg male-sterile are not fulfilled.

The inbred line TxGJ39, when used as male parent in a cross with 4632^{Vg}, did not restore fertility. One F_1 plant out of five grown in the greenhouse did shed pollen, and was selfed. The F_2 progeny from this plant were all sterile or only partially fertile. An F_1 population of 107 plants was grown in the field in 1953. None of the plants were classified as fertile. It is very possible that the plant which was selfed in the greenhouse would not have shed pollen under field conditions.

The F_1 plants of the cross 4632^{Vg} x K64 were sterile, both in the greenhouse and in the field.

Seven plants of the F_1 from 4632^{Vg} x K55 were grown in the greenhouse. Six were sterile and one partially fertile. A total of 109 F_1 plants were grown in the field, noneof which shed pollen.

When 4620^{R} was crossed with TxGJ39, the F_1 plants were fertile in the greenhouse, but not all were fertile in the field. The material

grown from the first two planting dates included male-sterile, partially fertile, and fertile plants. All plants of the third (last) planting date were fertile. The cross 4620^{R} x TxGJ39 is not suited for genetic study because of the apparent environmental effect on expression of male-sterility.

Inbred lines K64 and K55, when used as male parents in crosses with 4620^{R} did not restore fertility in the F_1 generation.

In the 4632^{Vg} x Ky 39 cross four out of seven F_1 plants grown in the greenhouse were fertile. In the field five out of seven were fertile. In the cross 4632^{Vg} x Ky39 all of 58 progeny were fertile when grown in the field. This suggests that Ky 39 was homozygous for the restorer gene or genes, and that the 4632^{Vg} parent used was heterozygous either at the same locus or loci concerned in Ky 39, or that the female parent was heterozygous for modifiers which when complemented by the genotype of Ky39 resulted in expression of male-sterility.

One of the fertile 4632^{Vg} x Ky 39 plants was selfed and the F_2 progeny grown in the field. A fertile F_1 plant was backcrossed to the male-sterile parent, and a male-sterile F_1 plant backcrossed to the Ky39 parent. The F_2 was grown at two different planting dates. Chi square in a test for independence of effects of dates was significant with a P value of .02 so one might conclude that the expression of male-sterility in the two F_2 populations was affected by environment. The classifications of the segregating generations for this cross and with Ky21 are given in Table 31.

Nearly all plants were fertile in the F_2 and in the backcross to the

Table 31. Number of plants within each male-sterile or male-fertile class for segregating generations of crosses involving the Vg source of cytoplasmic male-sterility, at designated planting dates.

Class[a]	4632^{Vg} x Ky39 F_2		(4632^{Vg} x Ky39) x Ky39	4632^{Vg} x (4632^{Vg} x Ky39)	4632^{Vg} x Ky21 F_2		(4632^{Vg} x Ky21) x Ky21	4632^{Vg} x (4632^{Vg} x Ky21)
	May 9	May 26			May 9	May 26		
	Number of plants							
0	-	-	-	-	7	2	-	16
1	-	3	-	1	4	3	-	4
2	1	-	-	-	2	2	-	4
3	-	-	-	-	1	3	-	-
4	1	-	1	-	3	-	-	8
4pf	1	-	-	-	-	-	-	-
5	1	3	12	8	14	13	6	3
5pf	5	-	1	7	22	9	2	7
5f	93	37	28	61	144	49	17	104
Total	102	43	42	77	197	81	25	146

[a]Classes are the same as those described at the end of Table 29.

male-sterile parent. This again suggests the possibility of some type of selective fertilization or of a part of the pollen not functioning in the F_1 generation. In the backcross of a male-sterile F_1 plant to Ky39 more than one-half of the plants were fully fertile. It may be that an important modifier had been contributed by the 4632^{Vg} parent and was present in the male-sterile F_1, being epistatic to the restorer gene or genes from Ky39. When backcrossed to Ky39, if a single modifier gene was involved, one would expect about one-half fertile plants and one-half male-sterile plants. The data are not conclusive.

All F_1 plants of 4632^{Vg} x Ky21 grown in the greenhouse were fertile. Fourteen out of 16 grown in the field were fertile. No explanation for the presence of the two male-sterile F_1 plants is attempted, other than considering the possibility that they represent contaminant pollinations. All of 58 4652^{Vg} x Ky21 plants were fertile in the field.

The possibility that expression of male-sterility in the F_2 was to some degree affected by environment is indicated by the almost significant chi square value in the test for independence of date of planting effects. The P value obtained was .07. The system of classification given in Table 31 is not adequate for recognition of phenotypes, so definite ratios are not indicated. Again the fertile classes are predominant in the F_2 and both backcrosses, indicating that there may be some chance of identification of genotype upon examination of pollen, or that selective fertilization may have occurred. One would expect a fertile progeny in a backcross to Ky21, since Ky21 acted as a fertility restorer in the F_1. It seems that there must be some segregation of modifiers independent of

Table 32. Number of plants within each male-sterile or male-fertile class for segregating generations of crosses involving the Reid source of cytoplasmic male-sterility, at designated planting dates

Class[a]	4620^R x Ky39 F_2	(4620^R x Ky39) x Ky39	4620^R x (4620^R x Ky39)	4620^R x Ky21 F_2		(4620^R x Ky21) x Ky21	4620^R x (4620^R x Ky21)
				May 9	May 26		
			Number of plants				
0	1	-	2	0	1	1	6
1	2	-	3	0	4	-	-
2	-	-	1	-	-	-	1
3	1	-	1	2	-	-	3
4	2	-	-	1	1	-	-
4pf	-	-	-	-	-	-	-
5	13	9	2	10	4	7	12
5pf	19	3	-	10	2	3	5
5f	84	10	54	94	47	24	47
Total	122	22	63	117	59	35	74

[a]Classes are the same as those described at the end of Table 29.

the restorer gene or genes. These modifiers must have been introduced through the male-sterile parent, and in the right combination interact with the genotype of Ky21 to produce male-sterility.

The F_1, F_2, and both backcross populations of 4620^R x Ky39 are very similar to the respective populations of 4620^R x Ky21 (Table 32). Both F_1 populations were fertile and in each F_2 most plants were classified as fertile. The F_2 involving Ky21 was grown at two different planting dates. Significant chi square in a test for independence of date of planting effects indicates variability in classification under different environmental conditions. No clear cut segregation is evident in any of the populations, but there again is a trend toward high frequency of fertile plants in the F_2 populations and in the backcrosses. As was stated in the presentation of the 4632^{Vg} x Ky21 data, examination of pollen may prove informative.

DISCUSSION

Within the limits of fertility differential test crosses made, the U.S.D.A., Brazilian, Vg, and Reid sources of cytoplasmic male-sterility were not greatly different in the cytoplasmic-genotypic interactions involved. No detailed accounts on genetic studies have been reported in any of these forms of male-sterility, but preliminary investigations into the manner of inheritance of the Vg and of the Reid sources suggest a similar mechanism involved in each of the two types. An effective method of testing the degree of likeness of these four sources of cytoplasmic male-sterility would be to introduce each of them into a common inbred line by means of a backcrossing procedure. A minimum of five backcrosses to the recurrent inbred line parent in each case is suggested. A critical genetic investigation could be carried out by crossing the four male-sterility sources, now in a common inbred line background, with an effective fertility restorer, such as Ky21 or Ky39, and a study made of the segregating generations. The line or lines selected for use as pollen parents should have been maintained strictly by selfing on an individual ear basis, to insure homozygosity. Any differences between the four crosses and their respective segregating generations, other than those environmentally influenced, may then be attributed to the cytoplasmic-genotypic interaction specific to each source of cytoplasmic male-sterility.

The Texas form of male-sterility was distinctly different from each of the other seven sources. Lines which restore fertility to the Texas

type in several cases did not act as fertility restorers to the other male-sterile forms. When Ky39 was used as the pollen parent in a cross with a Texas male-sterile line a completely male-sterile progeny was produced. Ky39 restored fertility to all lines involving other sources. Reaction of lines K64 and M14 specific to the Texas male-sterility was particularly noted. Nearly all individual plants of the F_1 populations involving these two lines were partially fertile. Very few if any partially fertile plants occurred in the F_1 populations involving K64 and M14 with the other types of male-sterility.

The Kys form of male-sterility was unlike any of the others included in this investigation. All fertility differential lines restored fertility when used as pollen parents in crosses with this source.

The two types of male-sterility represented by single crosses are difficult to compare with the others. A more effective method of testing each with the series of fertility differentials would be to use the lines J.C.33-16 and M1984 as pistillate parents, instead of the male-sterile single crosses. On the basis of tests made, the two types appear to differ from each other and both are unlike any of the other sources of male-sterility observed.

The modifier background of an inbred line converted to the U.S.D.A. type of cytoplasmic male-sterility appears to influence the degree of expression of male-sterility to a greater degree than for lines converted to the Texas source. Lines with the U.S.D.A. type of sterility in common did not react the same way in crosses with several of the tester differentials. However, all crosses did not reach anthesis at the same time and it is difficult to separate environmental response from nuclear-

cytoplasmic interaction. For example, the first two planting dates of A^S, $WF9^S$, and $A158^S$ all crossed with TxGJ39 were the same in expression of the male-sterile character. On the third date the F_1 populations involving $WF9^S$, a late maturing line, and $A158^S$, an early maturing line, consisted of partially fertile plants. In the same late date of planting, A^S, another early maturing line, produced an F_1 with no partially fertile plants. It seems that these lines differ in male-sterility response because of unlike genotypes, but how much of the variability may be attributed to environmental fluctuations is not known.

Crosses of the fertility differential stocks with lines converted to the Texas source were uniform in expression of the male-sterile character, except for those involving K64 and M14. Considering these exceptions, the particular inbred line involved in the cross seemed to have a direct effect on expression of male-sterility in the F_1 plants. Particularly in the comparison of $Tx203^T$ x M14 and $Tx61M^T$ x M14, a difference between the two crosses was maintained through each of the three planting dates, even though time of planting seemed to have considerable influence on degree of male-sterility expressed.

When the original cytoplasmic male-sterile stocks, except for J.C.33-16 x Mo2RF, were each planted at the three different dates, only the incompletely male-sterile single cross M1984 x M14 showed significant variability in expression of male-sterility. However, when these same male-sterile stocks were crossed with the series of fertility differential testers, a number of the F_1 populations proved to be markedly influenced in expression of the male-sterile character by date of planting. The single cross M1984 x M14 proved to be one of the male-steriles more susceptible to environmental influence on degree of pollen shedding when

crossed with the tester series. The U.S.D.A. and the Vg sources, and to a more limited extent 4620^R, seemed to be quite highly influenced in degree of anther exsertion through time of planting in several F_1 combinations. This would suggest that the expression of male-sterility, particularly in the case of some sources, is influenced by modifier genes, some of which very likely achieve penetrance only under certain environmental conditions. The environmental effect observed through growing the same material at different stages of maturity may be that of temperature, humidity, a combination of the two, or possibly day-length. The three planting dates chosen were approximately two weeks apart, so that more than one month intervened between the first and third dates. It is conceivable that length of day could have had some effect. Over that period of time temperature and humidity could well have had marked influence. In most instances the material of the last planting date showed the highest degree of anther exsertion, and if any pollen was shed, it was usually in the late planting. Some lines of the Texas source of male-sterility when combined with lines K64 and M14 showed most anther exsertion and partial fertility at the early planting date. Most crosses grown on the third date reached anthesis during a period when cooler temperatures prevailed, with relatively high humidity. These conditions may have been more favorable for exsertion of anthers and shedding of pollen than the considerably higher temperatures which were general about the time the first planting date material reached anthesis.

An experiment could be devised to test the possible effect of day-length on degree of expression of male-sterility. By controlling length

of time of exposure to daylight in a replicated experiment, using a designated number of different light periods, one could detect any noticeable influence on anther exsertion or pollen shedding.

Segregation for male-sterile and partially fertile or fertile plants in the F_1 generation of a cross between a cytoplasmic male-sterile line and a long-time inbred line can well be attributed to heterozygosity for important modifiers in the male-sterile parent. In some experiments reported (15)(34), such variability in expression of male-sterility in the F_1 has been explained by assuming that the inbred line used as male parent was heterozygous for fertility restoring genes. An established inbred line is apt to be more highly inbred than the male-sterile parent, and thus would be expected to be less heterozygous. In tests reported on in this paper no relic heterozygosity for fertility restoring genes was found in any of the individual Hy or C.I.7 plants outcrossed to male-sterile inbred lines, but in outcrosses made in the same manner to male-sterile single crosses several progenies were obtained which were segregating for male-sterile and partially fertile or fertile plants. The individual Hy and C.I.7 plants used were selected at random from five different sources of each line. In some cases a small number of plants were available for test but the results indicate that if a homozygous male-sterile parent is used there is no segregation for male-sterility in the F_1 population. The variable F_1 population produced from some 3-way crosses involving a male-sterile single cross as female parent suggests that segregation was due to heterozygostiy of modifiers in the female parent and not of fertility restoring genes in the pollen parent.

The comparative morphological study of the incompletely male-sterile

single cross M1984 x M14 with the fertile reciprocal cross M14 x M1984 indicated that at the time of meiosis the anthers of the former cross were slightly shorter and of smaller diameter than the anthers of the latter cross. Approximately eight days following meiosis a very marked depression occurred in the growth rate of the anthers in the incompletely male-sterile single cross, both in length and in diameter. At the same time there was a significant difference between pollen diameter in M1984 x M14 and pollen diameter in M14 x M1984, but further growth and development ceased in about one-half of the pollen grains in M1984 x M14. The remainder of the pollen continued to increase in diameter and in iodine staining capacity, but not at the same rate of development as was observed for the fertile cross M14 x M1984. At the time of anthesis there was a highly significant difference in diameter of normal staining pollen grains between the two crosses.

If the expression of incomplete male-sterility in M1984 x M14 was due to a single gene in conjunction with a specific cytoplasmic factor, as the data presented suggest, that gene must be pleiotropic in effect. The reduced growth and development of all anthers in tassels of M1984 x M14 (Ss) plants is apparently only one form of expression of the gene. Random distribution of the two alleles, S and s, to microspores is accomplished through meiosis, and apparently the S gene has a second effect, that of cessation of growth and development on the microspores. Those microspores bearing the S allele fail to complete normal development while the microspores with the s allele proceed to develop into functional pollen grains.

It does not seem logical to assume that the cytoplasmic factor concerned in the incompletely male-sterile single cross is particulate in nature. If it were, the particles presumably would be distributed at random through somatic cell division in the development of the plant and specifically the tassel. An occasional cell, with its derivitive cells, in all probability would be devoid of such particles, resulting in fertile sectors of the tassel, or even an occasional normal anther. No such plants were observed during the two seasons the single cross M1984 x M14 was grown. All F_1 plants were very uniform in degree of anther exsertion, in appearance of anthers, and in the very limited amount of pollen shed. If cytoplasmic particles were dispersed at meiosis to the microspores it seems that more variability among F_1 plants would be encountered in the ratio of functional pollen grains to those which are non-functional.

The effect of modifier genes is suggested by the fact that a number of F_2 plants were classified as other than fertile or intermediates as typified by the F_1 M1984 x M14 plants. Similar deviations from expectation were observed in the backcross populations. Apparently the male-sterile character was highly influenced through the expression of modifier genes. Significant chi squares for tests of independence between planting date effects in segregating generations of M1984 x M14 may indicate that the modifying factors have a threshold effect and attain penetrance only under specific environmental conditions. The expressivity of these modifiers may in turn be very dependent upon environment.

A point in favor of the possibility of a series of modifiers which affects the expression of sterility is the fact that the inbred lines M1984 and M14 are so different in degree of pollen production. M1984 is a line

which is highly variable in amount of pollen shed under different environmental conditions. At temperatures from 70° to 75°F. accompanied by favorable humidity, an abundance of pollen is shed, but under hot and dry climatic conditions this line produces very little pollen. Oftentimes the anthers dry up without the pores having opened. M14 is quite different in that under most temperature and humidity conditions prevalent in Iowa during midsummer this line is a very satisfactory pollen producer. On the basis of these observations it can be expected that the two lines differ at perhaps several loci which have some effect on the expression of sterility or fertility. That the lines are homozygous at all loci involved is evidenced by the extreme uniformity of the F_1 plants.

All classification of plants was based on tassel appearance. If the hypothesis is true that sterility results from a single major gene S, one effect of which is immediately expressed in the pollen, then a system of classification based upon examination of the pollen from each plant would be much more satisfactory. Thus the phenotype would probably not be masked by the modifier genes as was very likely the case in the system which was used. In any future study involving the cross M1984 x M14 it would be desirable to take tassel samples at the time of anthesis from individual plants of the F_2, reciprocal backcrosses to both parents, and available F_3 populations.

The two sources of cytoplasmic male-sterility, Vg and Reid, seemed to have a similar interaction of the cytoplasmic factor with the genotype of the six lines used as pollen parents in the genetic investigation. The hypothesis has been presented that a male-sterile plant is of the

genotype SS or Ss, accompanied by the necessary cytoplasmic factor, and that an inbred line which acts as a fertility restorer bears a dominant allele designated as S'. There appears to be some type of selective fertilization encountered when using a heterozygote of the genotype SS' as a male parent. Observation of pollen produced by an F_1 plant should determine whether or not it is a matter of only one type of pollen grain functioning. If there is no visible difference between genotypes of pollen grains, a test can be made in an attempt to establish the presence of a selective male gametophyte factor. A cross using the F_1 plant as the pistillate parent, with some homozygous inbred line which has no effect on the expression of sterility of the Vg or the Reid source as the male parent, should produce a segregating progeny. Presumably the pollen parent would carry no restoring genes and no modifier genes which might mask the phenotype of the progeny of this cross. The line would have to be the same genotype as the original sterile parent in respect to loci affecting the expression of male-sterility.

Explanation of minor differences in degree of fertility restoration to the Vg source and to the Reid source by the male parent lines used in the preliminary genetic study may lie in differences of modifier background in 4632^{Vg} and in 4620^{R}, or these differences may be in the cytoplasmic component of inheritance. Data presented do not furnish evidence on this question.

It seems possible that the Vg sterility could be introduced into a line such as M14 by means of a backcross procedure, so that in effect after several generations of backcrossing the new sterile line would

consist of the cytoplasm of the Vg nonrecurrent parent and the genic complement of the M14 recurrent parent. This would eliminate modifiers present in the Vg sterile stock which appear to be segregating in the study thus far made. If after several backcrosses to M14 the progeny are entirely sterile, it seems reasonable to conclude that M14 is free of modifiers. The same method could be used to convert M14 to a sterile M14 through use of the Reid source. Again, if after several backcrosses to M14 the progeny are entirely sterile, one may be justified in assuming that M14 is free of modifiers of the Reid sterility. After the $M14^{Vg}$ and $M14^{R}$ lines have been attained, a much more satisfactory genetic investigation could be made. A cross between $M14^{Vg}$ and an effective restorer line such as Ky21 could be compared with a cross between $M14^{R}$ and Ky21. If the F_1 plants were fertile, and the F_2 plants and backcrosses to each parent had the same respective ratios or distributions (within limits of statistical tests), it would indicate that the differences between 4632^{Vg} x Ky21 and 4620^{R} x Ky21 reported on in the present study were due to differences in the modifier background of the sterile stocks, and not in the cytoplasmic factor concerned.

SUMMARY

1. A total of eight cytoplasmic male-sterile corn types, each from a different source, was collected for purposes of comparison and evaluation. An indirect method, that of crossing each to a series of tester lines serving as fertility differentials, had to be adopted to test differences between types of male-sterility. Within the limits of test crosses made, the U.S.D.A., Brazilian, Vg, and Reid sources were not greatly different in the cytoplasmic-genotypic interactions involved. The Texas form of male-sterility was distinctly different from each of the other seven sources, as was also the Kys type. The two male-sterile single crosses, M1984 x M14 and J.C.33-16 x Mo2RF, are difficult to compare with the others, but on the basis of tests made the two types seem to differ from each other, and are unlike any of the other sources of male-sterility observed.

2. The modifier background of an inbred line converted to the U.S.D.A. type of cytoplasmic male-sterility appeared to influence the degree of expression of male-sterility more than in the case of lines converted to the Texas source.

3. In order to compare the various sources of cytoplasmic male-sterility in degree of environmental influence, each type was planted at three different dates, approximately two weeks apart. Single crosses involving each of the eight sources with the series of fertility differential tester lines were included in the date of planting experiment.

The single cross M1984 x M14 was the only one of the original cytoplasmic male-sterile stocks to show any significant variability in expression of male-sterility over the three planting dates. When the same male-sterile stocks were crossed with the series of fertility differential testers, a number of the F_1 populations proved to be markedly influenced in expression of the male-sterile character by date of planting.

4. An experiment was conducted to test individual plants of lines Hy and C.I.7 for presence of reported relic heterozygosity of fertility restoring genes. No evidence for heterozygosity at such loci was found in any of the Hy or C.I.7 plants outcrossed to male-sterile inbred lines. The presence of male-sterile and partially fertile plants, or in some instances male-sterile and fertile plants, in some F_1 populations produced by outcrossing Hy and C.I.7 plants to male-sterile single crosses suggests that segregation was due to heterozygosity of modifiers in the female parent, and not of fertility restoring genes in the pollen parent.

5. The comparative morphological study of the incompletely male-sterile single cross M1984 x M14 with the fertile reciprocal cross M14 x M1984 indicated that approximately eight days following meiosis a marked depression occurred in the growth rate of the anthers of the incompletely male-sterile single cross, both in length and in width. At the same period there was a significant difference between pollen grain diameter in the two crosses. Further growth and development ceased in about one-half of the pollen grains in M1984 x M14.

6. An hypothesis was formulated whereby the incomplete male-sterility of M1984 x M14 was explained as the expression of a single gene in

conjunction with a specific cytoplasmic factor. The gene is apparently pleiotropic in effect, one expression of which is the reduced growth and development of the anthers, and the second being the cessation of growth and development of microspores bearing the dominant allele. The genotype of an F_1 plant is represented as Ss. Upon selfing the F_1 only the male gametes with the s gene are functional, so that F_2 plants are either Ss or ss. Deviations from expectation encountered in the F_2 and backcross populations are attributed to the action of modifier genes, which apparently have a marked effect on the expression of the male-sterile character.

7. The Vg and the Reid sources of cytoplasmic male-sterility seemed to have a similar interaction of the cytoplasmic components with the genotype of six lines chosen as possible fertility restorers. A working hypothesis is presented, assuming that a male-sterile plant involving either the Vg or Reid type is of the genotype SS or Ss accompanied by the cytoplasmic factor, and that an inbred line which restores fertility in the F_1 generation bears a dominant allele designated as S'. Some type of selective fertilization is involved when using the heterozygote of the genotype SS' as the pollen parent.

8. Genetic investigations made are preliminary in nature. Further studies should include observation of pollen from individual plants of the F_1 and segregating generations, and a test for a selective male gametophyte factor by using the heterozygote as the pistillate parent in a cross with a non-fertility restoring line. More accurate genetic tests could be made if the male-sterile parents were inbred lines instead of heterozygous stocks.

LITERATURE CITED

1. Anderson, E. G. Maternal influence of chlorophyll in maize. Bot. Gaz. 76:411-418. 1923.

2. Bateson, W. and Gairdner, A. E. Male sterility in flax, subject to two types of segregation. Journ. Gen. 11:269-275. 1921.

3. Bauman, L. F. Results of further inheritance studies on the Kys type of cytoplasmic male sterility in corn. (Abstract.) Annual Meetings American Society of Agronomy. 1953.

4. Chittenden, R. J. Studies in variegation. II. Hydrangea and Pelargonium: with notes on certain chimerical arrangements which involve sterility. Journ. Gen. 16:43-61. 1925.

5. _____ and Pellew, Caroline. A suggested interpretation of certain cases of anisogeny. Nature. 119:10-11. 1927.

6. Demerec, M. A second case of maternal inheritance of chlorophyll in maize. Bot. Gaz. 84:139-155. 1927.

7. East, E. M. Studies on self-sterility. IX. The behavior of crosses between self-sterile and self-fertile plants. Genetics. 17: 175-202. 1932.

8. Foskett, R. L. Nature and inheritance of male-sterility in the onion variety, Scott County Globe. Unpublished Ph.D. thesis. Ames, Iowa, Iowa State College Library. 1952.

9. Gabelman, W. H. Reproduction and distribution of the cytoplasmic factor for male sterility in maize. Proc. Nat. Acad. Sci. 35:634-640. 1949.

10. _____. Genetic control of the cytoplasmic factor for male-sterility in maize. (Abstract.) Genetics. 37:582-583. 1952.

11. Gairdner, A. E. Male sterility in flax. II. A case of reciprocal crosses differing in F_2. Journ. Gen. 21:117-124. 1929.

12. Goulden, C. H. Methods of statistical analysis. 2nd ed. N. Y., John Wiley and Sons, Inc. 1952.

13. Johnson, T. Variation and the inheritance of certain characters in rust fungi. Cold Spring Harbor Symposia on Quantitative Biology. 11:85-93. 1946.

14. Jones, D. F. The interrelation of plasmagenes and chromogenes in pollen production in maize. Genetics. 35:507-512. 1950.

15. _____ and Mangelsdorf, P. C. The production of hybrid corn seed without detasseling. Conn. Agr. Exp. Sta. Bul. 550. 1951.

16. Jones, H. A. and Clark, A. E. Inheritance of male-sterility in the onion and the production of hybrid seed. Proc. Am. Soc. Hort. Sci. 43:189-194. 1943.

17. Josephson, L. M. and Jenkins, M. T. Male sterility in corn hybrids. Journ. Am. Soc. Agron. 40:267-274. 1948.

18. Karper, R. E. Maternal inheritance of chlorophyll in sorghum. Journ. Hered. 25:49-54. 1934.

19. Kempthorne, Oscar. The design and analysis of experiments. N. Y., John Wiley and Sons, Inc. 1952.

20. Lewis, D. Male-sterility in natural populations of hermaphrodite plants. New Phytologist. 40:56-63. 1941.

21. L'Heritier, P. The CO_2 sensitivity problem in Drosophila. Cold Spring Harbor Symposia on Quantitative Biology. 16:99-111. 1951.

22. Lindegren, C. C. Mendelian and Cytoplasmic inheritance in yeasts. Ann. Miss. Bot. Garden. 32:107-123. 1945.

23. Michaelis, P. Interactions between genes and cytoplasmic in Epilobium. Cold Spring Harbor Symposia on Quantitative Biology. 16:121-129. 1951.

24. Myers, W. M. Effects of cytoplasm and gene dosage on expression of male sterility in Dactylis glomerata. (Abstract). Genetics. 31:225-226. 1946.

25. Nutman, P. S. Nuclear and cytoplasmic inheritance of resistance to infection of nodule bacteria in red clover. Heredity. 3:263-291. 1949.

26. Owen, F. J. Cytoplasmically inherited male-sterility in sugar beets. Journ. Agr. Res. 71:423-440. 1945.

27. Preer, J. R. Some properties of a genetic cytoplasmic factor in Paramecium. Proc. Nat. Acad. Sci. 32:247-253. 1946.

28. Randolph, L. F. Cytology of chlorophyll types of maize. Bot. Gaz. 74:337-375. 1922.

29. Rhoades, M. M. Cytoplasmic inheritance of male sterility in Zea Mays. Science. 73:340-341. 1931.

30. _____. The cytoplasmic inheritance of male sterility in Zea Mays. Journ. Gen. 27:71-93. 1933.

31. _____. Genic induction of an inherited cytoplasmic difference. Proc. Nat. Acad. Sci. 29:327-329. 1943.

32. _____. Plastid mutations. Cold Spring Harbor Symposia on Quantitative Biology. 11:202-207. 1946.

33. _____. Gene induced mutation of a heritable cytoplasmic factor producing male sterility in maize. Proc. Nat. Acad. Sci. 36:634-635. 1950.

34. Rogers, John S. and Edwardson, John R. The utilization of cytoplasmic male-sterile inbreds in the production of corn hybrids. Agron. Journ. 44:8-13. 1952.

35. Sass, John E. Botanical microtechnique. 2nd ed. Ames, Iowa, The Iowa State College Press. 1951.

36. Schwartz, Drew. The interaction of nuclear and cytoplasmic factors in the inheritance of male sterility in maize. Genetics. 36: 676-696. 1951.

37. Sonneborn, T. M. Gene and cytoplasm. I. The determination and inheritance of the killer character in variety 4 of Paramecium Aurelia. Proc. Nat. Acad. Sci. 29:329-338. 1943.

38. _____. Gene and cytoplasm. II. The bearing of the determination and inheritance of characters in Paramecium Aurelia on the problems of cytoplasmic inheritance, Pneumococcus transformations, mutations and development. Proc. Nat. Acad. Sci. 29: 338-343. 1943.

39. Spiegelman, S. The particulate transmission of enzyme-forming capacity in yeast. Cold Spring Harbor Symposia on Quantitative Biology. 16:87-98. 1951.

40. Srb, Adrian M. and Owen, Ray D. General genetics. San Francisco, W. H. Freeman and Company. 1952.

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APPENDIX

Table 33. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14, at sampling date number one. Stage of microsporocytes or microspores is included.

M1984 x M14								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microsporocyte or microspore[d]	Stage of development of microsporocyte or microspore
1	1	90	38.3	19.3	15.3	10.3	10	pachytene
	2	80	33.3	17.3	16.0	11.0	10	"
	3	82	31.0	16.3	13.0	9.7	10	"
	4	80	34.3	17.7	14.0	10.0	10	"
	5	90	36.6	19.0	14.3	12.3	10	"
4	1	90	38.7	19.3	16.7	10.3	9	"
	2	102	42.7	23.0	14.3	10.3	5	within quartet
	3	102	42.7	22.3	15.0	10.7	5	"
	4	102	42.0	24.0	15.0	10.7	5	"
	5	91	40.7	20.3	15.0	10.3	10	1st anaphase
5	1	86	34.7	16.0	14.0	9.7	10	pachytene
	2	82	38.7	18.7	15.0	10.0	10	"
	3	95	41.3	23.3	14.7	10.3	5	within quartet
	4	85	38.7	19.7	15.3	11.0	10	pachytene
	5	90	40.7	21.7	15.7	10.3	5	within quartet

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

Table 33. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microsporocyte or microspore	Stage of development of microsporocyte or microspore
6	1	81	33.7	15.0	14.3	10.3	10	pachytene
	2	92	42.0	23.0	15.0	11.0	10	diak. & 1st meta.
	3	85	39.3	21.0	15.3	11.0	10	pachytene
	4	92	40.0	19.3	15.0	11.7	10	"
	5	86	41.0	23.3	16.0	12.3	10	dipl. & diak.
7	1	85	40.0	22.0	16.3	11.7	10	diak. & 1st meta.
	2	83	35.0	18.7	14.3	11.7	10	pachytene
	3	82	39.3	20.3	15.3	10.3	10.5	diakineses
	4	85	36.7	19.3	15.0	12.3	10	pachytene
	5	80	34.0	18.7	14.7	10.7	10	"
8	1	72	32.3	16.0	13.0	9.7	10	"
	2	70	31.7	16.3	14.0	10.3	10	"
	3	82	36.7	18.0	14.3	11.0	10	pach. & dipl.
	4	70	30.7	15.7	12.0	10.0	10	pachytene
	5	70	33.3	18.0	14.0	11.3	10	"
9	1	65	28.0	13.3	12.0	9.0	10	"
	2	66	30.7	15.0	12.3	10.0	10	"
	3	60	31.7	17.0	13.0	10.0	10	"
	4	76	33.3	17.0	14.0	11.3	10	"
	5	75	30.7	14.3	12.7	10.3	10	"
10	1	100	40.3	20.3	14.3	11.3	10	diakinesis
	2	95	40.0	20.3	15.0	10.3	10	diak. & 1st meta.
	3	75	34.7	17.7	13.0	10.0	10	pachytene
	4	95	35.3	19.0	13.7	10.3	10	"
	5	95	36.7	20.7	14.3	10.0	10	"

Table 33. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microsporocyte or microspore	Stage of development of microsporocyte or microspore
11	1	66	28.0	11.7	11.0	8.7	10	pachytene
	2	70	31.3	15.3	13.7	10.3	10	"
	3	80	36.0	19.0	13.7	11.0	10	"
	4	76	35.0	18.0	14.0	10.3	10	"
	5	76	32.0	16.0	13.3	10.0	10	"
12	1	80	35.0	17.3	13.7	10.0	10	"
	2	90	35.3	18.7	13.3	10.3	10	"
	3	90	40.0	21.7	13.3	10.3	10	"
	4	85	38.3	20.0	14.7	10.7	10	pach. & dipl.
	5	98	37.0	18.7	14.0	11.0	10	"
13	1	68	34.0	17.3	13.3	10.0	10	pachytene
	2	75	33.0	16.0	12.3	10.0	10	"
	3	80	35.3	17.7	13.0	10.3	10	"
	4	68	30.7	15.0	12.3	10.0	10	"
	5	80	35.3	18.7	14.0	11.0	10	"
14	1	82	39.0	20.0	14.3	10.3	10	1st anaphase
	2	80	38.0	17.7	15.0	10.3	10	dipl. & diak.
	3	85	39.3	21.3	14.7	11.0	5	within quartet
	4	75	33.3	15.7	13.0	9.7	10	pachytene
	5	75	34.3	9.0	14.3	7.7	10	dipl. & diak.

Table 34. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the fertile reciprocal cross M14 x M1984, at sampling date number one. Stage of microsporocytes or microspores is included.

M 14 x M1984								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microsporocyte or microspore[d]	Stage of development of microsporocyte or microspore
1	1	80	32.3	14.0	15.7	12.3	9.5	pachytene
	2	84	38.3	19.0	20.3	14.3	4.5	within quartet
	3	80	35.7	17.3	17.3	12.7	8.5	2nd anaphase
	4	86	35.7	18.7	17.3	12.0	8.5	"
	5	80	37.7	21.7	17.0	12.0	5.0	within quartet
2	1	85	42.0	22.3	18.0	16.3	4.5	"
	2	80	40.3	24.0	19.3	15.0	8.5	2nd anaphase
	3	87	45.0	26.0	19.0	14.0	4.5	within quartet
	4	90	40.3	22.3	19.0	13.3	5.0	"
	5	92	42.7	25.0	15.3	13.7	4.5	"
4	1	100	42.3	23.3	20.3	12.3	5.6	free microspores
	2	105	41.7	22.0	20.7	14.7	5.0	"
	3	93	43.0	23.3	20.3	13.3	4.5	within quartet
	4	86	37.3	21.0	19.3	14.0	5.0	"
	5	80	35.7	19.0	16.3	12.3	8.9	1st anaph. & meta.

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

Table 34. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microsporocyte or microspore	Stage of development of microsporocyte or microspore
5	1	100	44.3	25.7	20.3	13.0	5.0	within quartet
	2	97	44.3	23.0	20.0	11.3	5.0	"
	3	90	45.0	24.7	18.3	13.7	5.5	free microspores
	4	90	40.0	20.0	19.3	11.0	5.0	within quartet
	5	95	43.0	23.7	19.3	11.3	5.5	free microspores
6	1	80	38.3	15.7	17.7	10.7	4.5	within quartet
	2	87	37.0	18.3	18.3	11.7	5.0	free microspores
	3	88	41.0	19.3	20.0	14.3	5.5	"
	4	90	38.0	19.3	19.0	12.3	5.5	"
	5	80	38.3	16.3	16.3	12.3	5.0	within quartet
7	1	70	28.3	13.0	11.3	10.3	10.0	pachytene
	2	65	29.0	14.7	14.3	10.7	10.0	"
	3	62	27.0	12.0	12.0	9.7	9.5	"
	4	66	27.0	11.3	12.0	10.0	9.5	"
	5	60	29.0	12.0	13.0	10.3	10.0	"
8	1	83	39.7	21.0	17.3	11.3	5.0	within quartet
	2	90	45.7	24.3	18.3	14.0	5.5	free microspores
	3	95	43.7	26.0	19.0	13.0	5.5	"
	4	84	41.3	21.3	16.3	12.3	5.0	within quartet
	5	86	39.0	18.0	16.3	11.7	9.0	free microspores
9	1	75	30.0	17.0	16.0	10.0	10.5	pachytene
	2	75	33.3	19.0	17.0	11.3	10.0	"
	3	64	28.7	14.3	14.3	12.0	10.5	"
	4	90	36.7	21.0	16.7	12.0	9.5	"
	5	92	33.3	20.0	16.0	10.0	9.5	"

Table 34. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microsporocyte or microspore	Stage of development of microsporocyte or microspore
10	1	72	28.3	14.3	14.0	10.7	10.0	pachytene
	2	84	35.0	17.7	15.0	9.3	9.5	"
	3	92	36.0	19.7	19.0	12.3	9.0	diplotene
	4	86	37.0	20.7	17.0	11.0	7.5	1st meta. & anaph.
	5	72	31.0	15.7	15.0	12.0	10.0	pachytene
11	1	82	34.0	17.3	15.0	10.0	10.0	pach. & dipl.
	2	80	33.3	15.0	14.3	11.7	10.0	"
	3	80	32.0	15.0	14.0	12.0	10.0	"
	4	75	33.0	17.7	15.0	13.0	10.0	"
	5	76	30.3	13.7	13.7	11.3	10.0	pachytene
12	1	76	34.3	17.3	14.3	10.7	10.0	"
	2	78	35.0	16.7	15.3	10.3	10.0	"
	3	85	38.7	22.3	17.3	12.7	9.5	1st anaphase
	4	82	38.3	19.7	15.7	11.3	8.5	1st meta. & anaph.
	5	75	34.0	16.3	15.3	11.3	10.0	pachytene
13	1	85	44.0	26.7	20.0	16.0	5.0	within quartet
	2	95	41.3	19.0	17.0	10.7	5.0	"
	3	90	39.3	22.0	16.3	10.6	5.0	"
	4	80	39.3	19.0	17.3	11.3	5.0	"
	5	85	42.0	24.3	19.3	11.3	5.0	"

Table 35. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14, at sampling date number two. Stage of microspores is included.

M1984 x M14

Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore[d]	Stage of development of microspore
1	1	110	47.3	30.0	19.0	13.3	5.0	free microspores
	2	115	46.7	29.7	17.7	13.0	5.0	"
	3	110	46.7	29.3	16.7	12.0	5.0	"
	4	115	46.0	23.3	17.0	10.7	5.5	"
	5	110	44.3	26.3	17.0	12.7	5.5	"
4	1	125	58.0	39.0	20.0	14.3	9.0	"
	2	110	58.3	40.3	19.3	15.0	8.6	"
	3	145	61.0	43.3	19.7	14.7	8.6	"
	4	110	61.0	39.7	20.3	15.0	8.9	"
	5	120	54.0	39.0	20.0	15.3	8.0	"
5	1	125	49.7	37.7	16.7	15.0	8.2	"
	2	115	44.3	31.3	19.7	13.0	8.9	"
	3	125	57.0	36.0	20.0	15.0	8.2	"
	4	120	49.7	27.3	18.3	12.0	9.5	"
	5	110	49.7	30.0	19.0	12.7	9.0	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

Table 35. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
6	1	125	55.0	37.0	19.0	14.0	10.0	free microspores
	2	115	42.7	30.7	18.0	12.7	7.8	"
	3	125	51.0	35.7	18.0	13.3	8.5	"
	4	145	55.0	41.0	19.7	15.7	8.5	"
	5	115	50.0	36.7	18.7	14.0	7.0	"
7	1	125	56.0	38.7	19.3	14.7	10.0	"
	2	120	45.7	27.7	15.0	13.7	5.2	"
	3	130	55.7	28.3	20.0	12.7	8.5	"
	4	120	41.7	31.7	19.3	13.7	8.0	"
	5	110	51.0	34.7	18.7	14.3	8.5	"
8	1	120	51.7	31.3	19.0	13.7	9.0	"
	2	115	51.3	31.3	19.0	13.0	10.0	"
	3	120	54.0	37.7	19.0	14.3	10.0	"
	4	125	52.7	32.3	19.3	14.7	9.5	"
	5	110	49.3	27.3	20.0	11.7	8.5	"
9	1	125	58.7	42.3	20.7	16.0	10.0	"
	2	125	54.7	37.7	19.7	14.7	10.0	"
	3	130	57.3	41.7	20.0	15.3	10.0	"
	4	130	52.3	36.7	16.3	14.3	10.0	"
	5	120	56.7	39.0	20.3	15.0	10.0	"
10	1	130	53.3	40.7	18.0	16.0	10.0	"
	2	138	60.7	44.0	21.0	16.0	10.0	"
	3	125	58.3	40.0	20.0	15.0	10.0	"
	4	140	60.7	43.0	20.7	16.0	10.0	"
	5	120	54.3	42.7	20.0	16.0	10.0	"

Table 35. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
11	1	130	55.7	36.3	20.0	15.0	9.5	free microspores
	2	143	56.3	40.7	20.0	14.7	9.5	"
	3	130	51.3	31.3	19.0	13.0	8.5	"
	4	130	51.0	38.3	18.0	13.3	8.5	"
	5	140	56.7	37.3	20.7	14.0	8.5	"
12	1	105	47.7	22.0	16.0	12.0	8.9	"
	2	130	55.3	35.0	20.0	13.7	8.9	"
	3	125	52.3	32.7	19.3	13.0	8.9	"
	4	145	56.0	37.0	20.0	15.0	9.5	"
	5	120	47.3	27.0	17.3	13.0	5.0	"
13	1	110	40.7	34.7	13.3	13.0	5.0	-----
	2	135	61.0	43.3	20.0	15.3	10.0	free microspores
	3	130	56.7	39.3	20.3	15.3	10.0	"
	4	130	59.3	39.7	20.3	15.0	10.0	"
	5	120	54.7	38.3	20.0	13.3	10.0	"
14	1	120	48.3	33.0	17.7	12.0	8.0	"
	2	120	53.7	35.0	18.0	14.3	9.5	"
	3	120	52.7	35.0	19.0	16.0	9.5	"
	4	115	48.7	32.0	19.3	13.3	8.5	"
	5	110	47.0	32.0	18.3	12.7	7.0	"

Table 36. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the fertile reciprocal cross M14 x M1984, at sampling date number two. Stage of microspores is included.

M14 x M1984								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore[d]	Stage of development of microspore
1	1	130	59.3	39.3	21.3	16.3	10.0	free microspores
	2	125	57.3	40.7	21.3	16.7	10.0	"
	3	125	59.0	41.0	22.3	16.0	9.0	"
	4	140	59.7	43.0	21.7	17.0	10.5	"
	5	120	59.0	40.3	21.3	17.3	10.0	"
2	1	135	54.3	36.0	21.0	15.7	10.0	"
	2	135	56.3	38.3	20.3	16.0	10.5	"
	3	135	56.0	40.7	21.0	16.0	10.5	"
	4	130	53.7	37.7	19.3	15.0	10.0	"
	5	135	58.7	40.7	21.7	17.3	10.0	"
4	1	130	58.0	38.7	21.3	15.7	10.0	"
	2	135	61.0	39.0	21.3	16.3	10.0	"
	3	125	57.3	37.3	21.7	15.0	10.0	"
	4	120	58.3	39.0	20.7	14.7	10.0	"
	5	130	56.3	40.0	22.0	16.3	9.5	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

Table 36. (Continued)

M14 x M1984								
Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
5	1	125	59.3	40.7	23.0	17.3	11.0	free microspores
	2	130	58.7	41.0	22.3	16.3	10.5	"
	3	135	60.0	40.3	22.3	18.7	10.0	"
	4	130	62.0	45.7	22.7	18.3	10.0	"
	5	115	62.6	44.3	22.0	19.0	10.5	"
6	1	130	59.3	43.3	21.0	17.0	10.5	"
	2	130	61.3	42.3	22.3	16.7	10.0	"
	3	135	61.0	44.3	21.7	17.3	10.5	"
	4	135	57.3	41.0	21.0	16.3	10.5	"
	5	125	59.0	42.0	22.7	18.3	10.0	"
7	1	130	60.3	42.7	23.0	16.7	10.5	"
	2	125	62.7	44.0	22.7	16.7	10.5	"
	3	135	62.7	41.7	21.7	16.0	10.5	"
	4	140	62.3	44.0	21.7	17.7	10.5	"
	5	140	62.7	46.7	21.7	17.3	10.5	"
8	1	125	61.0	44.0	21.7	17.3	10.5	"
	2	130	61.3	40.7	22.7	17.3	10.5	"
	3	125	59.0	41.3	22.7	16.7	10.5	"
	4	125	61.3	42.3	22.3	18.0	10.5	"
	5	130	61.0	42.3	21.7	16.3	10.0	"
9	1	120	50.7	35.0	20.3	15.0	9.5	"
	2	120	51.0	31.7	19.3	14.0	8.5	"
	3	120	54.3	33.0	19.7	14.3	9.5	"
	4	110	49.3	29.7	19.7	15.3	8.0	"
	5	110	55.0	31.3	20.7	14.0	9.5	"

Table 36. (Continued)

M14 x M1984								
Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
10	1	115	52.0	32.3	20.7	14.3	9.5	free microspores
	2	125	47.7	27.7	18.3	13.0	8.0	"
	3	125	49.3	31.3	20.0	13.0	9.5	"
	4	115	47.7	32.7	19.3	15.3	9.5	"
	5	115	47.0	29.3	20.0	13.0	8.5	"
11	1	110	52.0	46.3	18.7	18.3	9.0	"
	2	120	58.7	40.7	21.7	16.3	10.0	"
	3	120	54.7	38.0	20.7	15.0	10.0	"
	4	130	57.0	37.7	22.3	15.7	10.0	"
	5	130	55.3	34.7	20.3	17.0	10.0	"
12	1	120	46.7	29.0	20.0	13.0	8.0	"
	2	110	44.0	25.7	16.7	13.7	5.0	"
	3	110	45.3	30.0	19.0	13.7	8.0	"
	4	105	43.7	28.0	17.0	13.7	8.0	"
	5	105	43.0	22.0	18.3	11.7	5.0	"
13	1	135	63.3	44.0	21.3	17.7	10.5	"
	2	135	63.0	44.0	22.0	16.3	10.0	"
	3	140	62.0	44.7	21.7	18.0	10.0	"
	4	125	57.0	42.0	21.3	17.0	10.5	"
	5	130	63.0	44.3	22.0	18.0	10.0	"

Table 37. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14, at sampling date number three. Stage of microspores is included.

M1984 x M14								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore[d]	Stage of development of microspore
1	1	145	65.3	56.7	21.0	20.3	9.0	no stain
	2	145	64.7	53.7	21.7	18.7	9.5	"
	3	130	55.7	44.0	19.7	17.7	9.0	"
	4	135	66.0	30.7	22.7	11.3	9.0	"
	5	140	64.0	54.0	22.0	21.0	9.0	"
4	1	150	74.7	65.7	24.7	21.7	10.0	"
	2	140	70.3	60.3	23.0	20.3	9.5	"
	3	130	73.0	64.0	22.0	20.3	9.5	"
	4	145	70.3	60.0	22.3	22.0	9.5	"
	5	150	72.0	64.7	23.3	20.0	9.5	"
5	1	130	62.3	57.7	22.0	20.0	9.0	"
	2	130	58.3	57.7	20.7	19.3	9.0	"
	3	140	68.7	60.7	21.7	20.3	9.0	"
	4	125	52.7	34.7	20.0	11.0	8.5	"
	5	130	56.0	52.0	18.0	20.0	9.0	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

Table 37. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
6	1	130	59.0	47.0	23.3	18.0	9.0	no stain
	2	140	69.7	58.7	23.7	20.0	9.5	"
	3	140	68.0	68.7	24.3	20.0	9.5	"
	4	140	70.7	60.0	24.0	20.3	9.5	"
	5	140	70.3	60.7	23.0	21.0	9.0	"
7	1	125	58.3	45.0	21.3	19.0	9.0	"
	2	130	67.3	58.3	23.0	19.7	9.0	"
	3	125	63.3	55.7	20.3	20.0	9.0	"
	4	140	69.7	59.7	22.3	20.3	9.0	"
	5	120	65.0	53.7	22.3	19.3	9.0	"
8	1	130	73.7	61.3	23.7	20.7	9.5	"
	2	130	68.7	54.3	21.7	18.7	9.5	"
	3	130	68.7	57.3	23.7	20.0	9.0	"
	4	130	68.0	60.0	23.3	20.0	9.5	"
	5	130	63.7	56.7	22.7	20.0	9.5	"
9	1	110	64.7	54.3	23.0	20.0	10.0	"
	2	135	67.0	53.7	22.7	20.0	10.0	"
	3	120	66.3	52.3	22.0	18.3	10.0	"
	4	130	65.0	53.3	22.7	18.7	9.5	"
	5	130	67.0	56.3	21.7	19.7	10.0	"
10	1	130	56.3	60.3	22.0	22.3	9.5	"
	2	135	72.3	62.0	24.7	21.7	9.5	"
	3	140	68.3	60.0	23.7	20.0	10.0	"
	4	130	66.7	58.7	22.7	21.0	10.0	"
	5	150	72.7	62.3	23.7	21.0	9.5	"

Table 37. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
11	1	125	70.0	59.3	24.7	20.0	10.0	no stain
	2	125	70.0	57.3	22.7	19.7	9.5	"
	3	140	68.7	56.7	24.0	20.0	10.0	"
	4	130	70.7	61.3	22.7	20.0	9.5	"
	5	130	68.0	57.0	25.0	19.7	9.5	"
12	1	135	65.0	49.3	21.7	18.7	9.5	"
	2	125	67.7	50.3	22.7	19.7	9.5	"
	3	135	63.0	47.3	21.3	18.3	9.5	"
	4	130	64.0	44.7	21.3	17.0	9.5	"
	5	140	61.3	45.7	22.3	19.0	9.5	"
13	1	125	52.0	57.3	20.0	20.3	9.0	"
	2	135	64.0	58.3	21.3	19.7	9.5	"
	3	135	70.3	60.3	23.3	20.7	9.5	"
	4	125	61.0	57.7	23.0	21.0	9.5	"
	5	125	65.7	57.7	22.0	20.3	9.5	"
14	1	130	65.3	54.3	22.0	18.7	10.0	"
	2	125	62.7	52.0	21.0	18.7	9.5	"
	3	130	63.0	48.3	22.0	19.0	10.0	"
	4	125	61.7	45.7	22.3	18.7	10.0	"
	5	120	54.7	44.7	21.0	17.7	9.5	"

Table 38. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the fertile reciprocal cross M14 x M1984, at sampling date number three. Stage of microspores is included.

M14 x M1984								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore[d]	Stage of development of microspore[e]
1	1	130	72.7	60.7	23.7	23.0	10.0	25
	2	140	72.7	61.3	23.7	20.7	10.0	"
	3	145	75.7	62.0	24.3	23.3	10.0	50
	4	140	68.0	58.0	23.7	22.0	10.0	25
	5	135	74.0	59.7	25.7	20.7	10.0	"
2	1	135	79.0	63.3	26.0	22.7	10.0	"
	2	140	76.0	65.0	28.0	23.0	10.0	"
	3	140	75.0	61.7	27.7	22.3	10.0	10
	4	140	69.0	64.0	24.3	22.0	9.5	5
	5	120	65.3	48.7	23.0	19.3	9.5	"
4	1	130	72.7	60.3	24.3	21.3	10.0	25
	2	130	73.0	61.3	24.7	22.0	10.0	10
	3	125	74.7	59.0	24.3	19.3	10.0	75
	4	125	67.3	60.0	25.3	22.7	10.0	50
	5	120	73.3	55.0	24.7	21.0	10.0	10

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]One micrometer division equals 6.6 microns.

[e]All microspores partially stained with iodine solution, number given represents mean per cent of microspore area which stained.

Table 38. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
5	1	130	72.7	58.7	24.0	21.7	10.0	50
	2	120	72.7	59.3	25.7	20.0	10.0	90
	3	120	73.0	59.7	26.3	21.7	10.0	75
	4	120	71.7	58.3	28.7	20.7	10.0	50
	5	120	74.0	61.0	26.7	20.3	10.0	90
6	1	125	70.0	53.3	24.7	21.3	10.0	25
	2	120	70.3	54.0	25.3	20.0	10.0	11
	3	140	70.0	56.7	24.7	21.3	10.0	75
	4	120	68.0	53.3	27.0	21.3	10.0	"
	5	120	70.0	56.0	27.3	20.3	10.0	90
7	1	130	70.3	59.0	25.0	21.0	10.0	25
	2	130	71.0	61.0	23.7	21.0	10.0	"
	3	145	75.3	62.7	26.7	20.7	10.0	10
	4	135	73.3	60.3	25.3	20.3	9.5	5
	5	140	75.7	62.0	28.3	21.3	10.0	25
8	1	120	68.0	52.7	24.3	21.3	10.0	75
	2	125	73.7	60.3	26.3	21.3	10.0	25
	3	130	77.3	63.3	25.0	21.3	10.0	90
	4	130	75.3	58.0	27.0	20.3	10.0	50
	5	135	74.3	62.7	25.0	22.3	10.0	75
9	1	125	66.7	55.3	24.3	21.0	10.0	5
	2	130	61.7	54.3	23.3	20.3	9.5	"
	3	130	68.3	56.0	25.0	20.0	9.5	10
	4	130	64.3	54.0	26.0	21.3	9.5	5
	5	120	63.3	55.0	24.0	22.0	9.5	"

Table 38. (Continued)

M14 x M1984								
Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore	Stage of development of microspore
10	1	135	63.7	55.3	24.0	20.3	9.5	5
	2	135	65.7	56.3	24.0	20.7	9.5	10
	3	120	63.0	56.3	23.7	20.0	9.5	"
	4	125	63.3	53.7	22.0	20.0	9.5	"
	5	125	64.7	50.7	23.0	19.7	9.5	5
11	1	135	71.3	60.3	26.3	22.0	9.5	10
	2	125	68.3	59.0	24.3	21.3	9.5	"
	3	130	68.7	59.7	23.7	21.7	10.0	5
	4	135	65.0	50.0	23.0	20.0	10.0	"
	5	135	65.3	60.0	23.7	22.0	9.5	"
12	1	135	65.3	46.0	24.0	17.7	9.5	5
	2	120	62.0	39.3	21.7	19.3	9.5	"
	3	130	66.0	44.3	23.7	18.7	10.0	"
	4	130	65.3	40.7	24.7	20.7	10.0	"
	5	135	63.0	43.0	22.0	20.0	10.0	"
13	1	120	69.7	58.3	29.0	22.7	10.0	90
	2	120	71.0	59.0	28.0	22.0	10.0	"
	3	125	72.3	59.0	28.0	21.0	10.0	"
	4	120	70.7	59.3	25.3	21.3	10.0	75
	5	120	72.3	59.0	26.0	23.3	10.0	50

Table 39. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14, at sampling date number four. Stage of microspores or pollen is included.

M1984 x M14									
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore or pollen[d]		Stage of development of microspore or pollen[e]
1	1	130	72.0	63.7	26.0	23.0	10.5	9.5	33
	2	135	76.7	70.0	25.7	23.0	10.5	9.5	50
	3	130	72.7	67.3	25.3	23.3	10.5	9.5	33
	4	130	71.0	67.3	24.7	25.3	10.0	10.0	25
	5	125	73.7	66.3	23.7	24.0	10.5	9.5	50
4	1	130	75.3	70.0	25.0	21.7	11.0	9.5	50
	2	120	62.7	66.0	20.7	24.0	11.0	9.5	33
	3	130	75.7	68.7	26.3	22.7	11.0	9.5	75
	4	125	74.0	68.0	22.7	21.7	11.0	9.5	50
	5	125	73.7	68.0	30.0	21.7	11.0	9.5	"
5	1	120	64.7	57.7	24.7	22.0	10.5	9.5	33
	2	120	66.7	64.7	23.0	22.3	10.5	9.5	50
	3	125	67.7	66.0	23.3	22.7	10.5	9.5	"
	4	120	69.3	66.3	22.7	23.0	11.0	9.5	33
	5	130	71.0	66.7	23.7	22.3	11.0	9.5	50

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]First column represents mean diameter for normal microspores or pollen grains, second column for microspores or pollen grains which do not stain. One micrometer division equals 6.6 microns. The stage at which the microspore nucleus divided to form the male gametophyte was not determined.

[e]Per cent of microspores or pollen grains which stained in full.

Table 39. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen		Stage of development of microspore or pollen
6	1	140	80.0	74.7	25.0	24.0	10.0	9.5	25
	2	130	76.7	66.3	27.3	24.3	10.0	9.5	"
	3	125	69.3	67.0	23.3	23.0	11.0	9.5	33
	4	130	70.3	52.0	24.0	20.7	10.0	9.5	5
	5	130	71.7	63.0	25.3	24.0	10.0	9.5	75
7	1	130	72.7	71.0	25.7	21.7	11.0	10.0	50
	2	130	75.3	72.0	24.3	23.7	11.0	9.5	"
	3	135	73.3	68.7	26.7	25.0	11.0	9.5	"
	4	135	73.7	70.0	26.3	24.0	11.0	9.5	75
	5	125	72.3	69.0	25.3	21.7	11.0	9.5	"
8	1	120	69.0	63.0	24.3	23.0	11.0	9.5	50
	2	110	68.7	63.7	25.7	23.0	11.0	9.5	"
	3	125	72.7	65.0	24.3	23.3	11.0	9.5	"
	4	120	73.0	68.0	23.7	23.0	11.0	9.5	"
	5	110	66.7	63.0	23.7	22.0	10.5	9.5	"
9	1	120	68.0	66.0	23.7	23.7	11.0	9.5	33
	2	130	71.3	65.0	23.7	22.7	11.0	9.5	25
	3	120	73.3	66.7	24.0	22.7	11.0	9.5	33
	4	120	66.3	68.0	23.3	24.0	11.0	9.5	50
	5	120	63.3	68.3	23.3	23.3	11.0	9.5	25
10	1	125	75.7	73.0	25.0	22.7	11.0	9.5	50
	2	130	76.0	70.7	25.3	22.3	11.0	9.5	"
	3	125	75.0	68.3	24.7	23.0	11.0	9.5	33
	4	125	76.7	71.0	28.0	22.7	11.0	9.5	50
	5	135	76.7	70.0	23.7	23.3	11.0	9.5	75

Table 39. (Continued)

M1984 x M14									
Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen		Stage of development of microspore or pollen
11	1	130	67.7	67.0	25.0	22.0	11.0	9.5	50
	2	130	68.7	66.7	25.0	22.0	11.0	9.5	"
	3	140	72.3	66.0	25.0	23.0	11.0	9.5	"
	4	120	71.3	67.7	24.0	21.7	11.0	9.5	"
	5	130	68.0	62.3	25.3	21.7	11.0	9.5	"
12	1	125	73.0	65.0	22.7	21.7	11.0	9.5	"
	2	125	72.0	67.0	24.0	22.7	11.0	9.5	"
	3	125	72.3	63.3	22.7	21.0	11.0	8.5	"
	4	130	70.7	65.3	24.3	21.3	11.0	9.5	"
	5	125	71.7	64.3	24.0	20.7	11.0	8.5	"
13	1	130	74.3	68.7	26.0	23.0	11.0	9.5	"
	2	120	69.7	63.3	25.0	21.0	10.5	9.5	"
	3	130	71.7	66.0	23.3	22.0	10.0	9.5	"
	4	120	73.0	63.0	25.3	23.7	10.0	9.5	"
	5	135	72.3	67.7	25.3	22.7	11.0	9.5	"
14	1	120	61.7	52.0	27.7	20.0	9.5	9.5	5
	2	120	68.3	55.3	22.7	20.0	10.0	9.5	33
	3	120	67.3	64.3	24.0	23.3	10.0	9.5	50
	4	125	69.3	64.7	23.7	23.0	11.0	9.5	"
	5	120	61.7	64.0	22.7	22.3	10.0	8.5	"

Table 40. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the fertile reciprocal cross M14 x M1984, at sampling date number four. Stage of microspores or pollen is included.

M14 x M1984								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore or pollen[d]	Stage of development of microspore or pollen[e]
1	1	140	86.7	78.7	32.7	27.7	11.0	99
	2	125	83.7	73.7	34.0	30.7	11.0	"
	3	135	87.3	87.0	33.0	33.7	11.5	"
	4	130	87.3	75.0	37.0	28.3	11.5	"
	5	135	86.3	74.3	34.7	32.3	12.0	"
2	1	135	84.0	71.0	38.7	23.3	10.5	"
	2	140	80.0	70.7	48.3	26.0	10.5	"
	3	135	80.0	69.3	40.0	24.0	10.5	"
	4	130	81.7	70.0	34.3	25.0	10.5	"
	5	120	80.7	61.7	35.0	25.7	10.5	95
4	1	120	80.3	67.3	36.0	31.7	11.0	99
	2	125	85.0	70.3	37.7	28.7	11.0	"
	3	125	78.3	67.7	36.7	24.0	11.0	"
	4	125	83.3	70.7	37.0	31.0	11.0	"
	5	130	85.7	71.7	40.0	28.7	10.5	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]First column represents mean diameter for normal microspores or pollen grains, second column for microspores or pollen grains which do not stain. One micrometer division equals 6.6 microns. The stage at which the microspore nucleus divided to form the male gametophyte was not determined.

[e]Per cent of microspores or pollen grains which stained in full.

Table 40. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen	Stage of development of microspore or pollen
5	1	125	88.3	76.3	45.3	41.7	10.5	99
	2	120	82.3	68.7	32.0	26.3	10.5	"
	3	125	87.3	76.3	38.7	31.0	11.5	"
	4	120	82.7	76.7	40.7	34.7	11.5	"
	5	135	91.7	78.0	43.3	42.3	11.5	"
6	1	110	80.0	72.7	40.0	28.3	10.5	"
	2	125	81.0	70.7	36.7	27.7	11.0	95
	3	110	80.0	68.3	38.7	31.7	11.0	99
	4	120	81.3	69.3	36.7	34.0	11.5	"
	5	120	81.3	74.3	38.3	36.0	11.5	"
7	1	125	87.7	73.0	36.7	28.7	10.5	99
	2	110	86.0	75.0	43.0	30.0	10.5	"
	3	130	86.7	73.3	40.0	31.3	10.5	"
	4	130	85.0	76.0	35.7	33.0	10.5	"
	5	125	80.0	67.0	37.7	31.7	11.5	95
8	1	110	84.0	73.7	37.0	33.3	11.5	99
	2	120	80.7	71.7	41.7	45.0	11.5	"
	3	120	81.3	70.0	48.3	32.0	11.5	"
	4	120	81.0	73.3	46.0	34.7	10.5	"
	5	120	88.3	76.7	50.0	41.0	11.5	"
9	1	110	70.3	64.0	35.0	28.0	9.5	95
	2	110	76.7	63.7	42.0	22.0	10.5	99
	3	120	81.0	68.7	43.0	23.7	10.5	"
	4	110	73.3	66.3	42.7	25.0	10.5	95
	5	115	79.3	65.0	45.0	23.7	10.5	99

Table 40. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen	Stage of development of microspore or pollen
10	1	125	79.0	71.7	50.0	31.0	10.5	99
	2	120	79.0	66.3	47.7	24.3	9.5	"
	3	130	78.0	62.0	47.7	25.0	10.5	"
	4	110	66.7	60.0	34.3	24.3	9.5	95
	5	135	84.3	71.7	43.3	40.0	10.5	99
11	1	125	77.6	68.0	39.3	28.7	10.5	"
	2	125	86.3	75.7	43.7	28.7	10.5	"
	3	120	83.7	74.0	48.3	27.7	10.5	"
	4	125	85.7	73.0	45.3	28.7	11.0	"
	5	130	83.3	64.3	47.7	31.0	11.0	"
12	1	120	84.3	71.0	42.7	25.0	11.0	99
	2	130	84.7	73.3	40.3	28.7	10.5	"
	3	125	83.3	65.3	46.7	26.0	10.5	"
	4	125	79.3	69.3	44.3	31.0	10.5	"
	5	125	83.0	67.7	47.7	26.0	10.5	"
13	1	125	85.3	73.0	46.7	32.0	11.0	"
	2	130	80.7	67.7	41.0	32.7	10.5	"
	3	135	88.7	75.0	47.7	32.7	11.5	"
	4	130	86.0	72.0	42.7	29.3	10.5	"
	5	130	87.3	71.3	47.7	32.0	11.0	"

Table 41. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the incompletely male-sterile single cross M1984 x M14, at sampling date number five. Stage of microspores or pollen is included.

M1984 x M14									
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore or pollen[d]		Stage of development of microspore or pollen[e]
1	1	125	74.7	68.0	27.7	26.0	12.5	10.0	50
	2	120	77.7	71.0	29.3	25.3	11.5	9.5	"
	3	135	74.3	72.0	26.7	27.0	11.5	9.5	33
	4	120	75.0	69.3	29.3	28.7	12.0	9.0	"
	5	130	76.0	64.7	29.3	27.0	11.5	9.0	50
4	1	120	72.0	69.7	30.3	26.0	12.0	9.0	"
	2	125	77.7	71.7	30.3	27.0	11.5	9.0	"
	3	125	79.3	70.3	29.3	29.3	12.0	9.0	"
	4	120	78.0	73.7	30.0	28.7	12.5	9.0	"
	5	125	68.3	68.0	28.7	26.0	12.0	9.5	"
5	1	120	74.3	29.3	26.3	26.3	10.5	9.5	"
	2	120	77.3	30.0	26.0	26.0	11.5	9.5	"
	3	120	73.0	34.3	30.3	30.3	11.5	9.0	"
	4	130	71.3	28.3	28.3	28.7	11.0	9.0	"
	5	125	71.0	34.3	34.3	27.7	10.5	9.0	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]First column represents mean diameter for normal microspores or pollen grains, second column for microspores or pollen grains which do not stain. One micrometer division equals 6.6 microns. The stage at which the microspore nucleus divided to form the male gametophyte was not determined.

[e]Per cent of microspores or pollen grains which stained in full.

Table 41. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen		Stage of development of microspore or pollen
6	1	130	79.3	66.0	32.7	26.7	11.5	9.0	50
	2	125	77.3	68.7	33.7	27.0	11.5	9.0	"
	3	130	75.0	69.7	32.7	28.0	11.5	9.0	"
	4	130	78.7	71.3	35.3	29.3	11.5	9.0	"
	5	120	76.0	71.0	32.7	26.0	11.5	9.0	33
7	1	120	70.0	69.0	34.3	25.0	11.5	9.5	50
	2	120	76.7	68.3	32.0	25.0	11.5	9.5	"
	3	120	76.7	67.0	31.0	26.0	11.5	9.5	"
	4	120	72.0	64.3	29.3	24.3	11.5	9.5	"
	5	130	74.0	68.7	37.7	28.7	11.5	9.5	"
8	1	120	71.3	69.0	27.7	29.3	10.5	9.5	33
	2	120	76.0	70.7	31.0	28.0	10.5	9.0	"
	3	125	76.7	68.7	30.3	26.0	12.0	9.5	50
	4	120	77.3	72.3	34.3	28.7	11.5	9.5	"
	5	120	73.7	70.3	32.0	27.7	10.5	9.5	"
9	1	130	78.7	70.7	30.0	26.7	11.0	9.5	"
	2	130	74.0	66.7	28.0	25.0	9.5	9.0	33
	3	120	77.7	73.3	33.0	27.0	10.5	9.0	50
	4	130	72.0	68.7	30.0	27.0	11.5	9.0	"
	5	120	77.3	69.3	32.0	27.7	11.5	9.0	"
10	1	130	80.7	71.0	28.7	28.0	11.5	9.0	"
	2	140	84.7	75.0	31.0	26.0	11.0	9.0	"
	3	140	80.7	76.3	31.0	27.0	11.0	9.0	"
	4	135	79.0	68.3	33.0	28.7	11.5	9.0	"
	5	125	74.3	65.7	31.0	27.0	11.5	9.0	"

Table 41. (Continued)

M1984 x M14

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen		Stage of development of microspore or pollen
11	1	135	76.0	70.7	31.0	26.0	11.5	9.5	50
	2	130	75.7	69.0	29.3	25.0	11.5	9.5	"
	3	125	75.0	68.0	29.3	27.0	11.5	9.0	"
	4	135	76.3	73.3	29.3	26.0	11.5	9.0	"
	5	120	74.0	66.7	30.3	24.3	11.5	9.0	"
12	1	125	74.7	64.3	28.7	24.3	11.5	9.5	"
	2	120	71.0	64.3	31.0	30.0	11.5	9.5	"
	3	120	71.7	62.0	33.0	28.0	11.5	9.5	"
	4	130	75.0	64.0	32.7	29.3	10.5	9.5	"
	5	120	74.3	64.3	29.3	25.0	11.5	9.5	"
13	1	120	75.0	64.3	27.0	25.3	11.5	9.5	"
	2	120	74.0	66.3	29.3	23.7	11.5	9.5	"
	3	120	71.3	64.7	28.0	23.0	12.5	9.5	"
	4	125	69.0	62.3	31.0	24.3	11.5	9.5	"
	5	120	72.7	65.7	30.0	25.0	11.5	9.5	"
14	1	125	73.0	61.7	29.3	25.3	11.5	9.5	"
	2	120	75.3	69.7	29.3	26.0	11.5	9.0	"
	3	125	75.0	67.7	30.0	27.0	11.5	9.0	"
	4	125	67.3	60.3	28.3	25.0	11.5	9.0	33
	5	120	71.3	61.3	29.3	26.0	11.5	9.0	50

Table 42. Individual plant measurements, in micrometer divisions, for glume length, anther length and width, and pollen diameter of the fertile reciprocal cross M14 x M1984, at sampling date number five. Stage of microspores or pollen is included.

M14 x M1984								
Plant number	Spikelet number	Glume length[a]	Mean anther length[b]		Mean anther width[c]		Mean diameter of microspore or pollen[d]	Stage of development of microspore or pollen[e]
1	1	130	104.7	81.3	38.7	38.3	13.0	99
	2	120	97.0	81.7	43.7	36.0	13.5	"
	3	130	100.3	88.3	46.0	37.0	13.5	"
	4	140	97.3	84.0	45.0	34.3	13.0	"
	5	130	100.0	89.3	46.0	40.3	13.5	"
2	1	130	95.3	81.0	47.0	39.3	12.5	"
	2	130	97.3	82.7	45.0	37.0	13.5	"
	3	135	95.0	86.7	48.0	37.0	13.0	"
	4	130	95.0	84.7	47.7	41.0	13.0	"
	5	135	91.7	84.0	44.3	32.7	12.5	"
4	1	120	87.3	80.3	39.3	38.7	12.5	"
	2	120	93.0	80.7	44.3	33.7	13.0	"
	3	115	93.7	80.0	48.3	32.7	13.0	"
	4	110	86.7	70.7	42.7	31.0	13.0	"
	5	115	93.0	79.0	37.7	34.3	14.0	"

[a]One micrometer division equals 70 microns.

[b]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 70 microns.

[c]First column represents mean measurement for pedicellate floret, second column for sessile floret. One micrometer division equals 28 microns.

[d]First column represents mean diameter for normal microspores or pollen grains, second column for microspores or pollen, grains which do not stain. One micrometer division equals 6.6 microns. The stage at which the microspore nucleus divided to form the male gametophyte was not determined.

[e]Per cent of microspores or pollen grains which stained in full.

Table 42. (Continued)

M14 x M1984

Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen	Stage of development of microspore or pollen
5	1	130	100.0	83.7	40.3	32.7	13.5	99
	2	120	94.7	81.0	39.3	35.0	13.5	"
	3	120	93.0	86.3	44.7	32.7	14.0	"
	4	120	98.0	86.3	39.7	34.3	13.5	"
	5	120	89.7	83.7	40.0	34.3	13.5	"
6	1	120	97.0	76.0	37.0	33.7	12.5	"
	2	120	99.7	76.3	42.7	32.0	12.5	"
	3	120	92.0	77.3	46.0	33.7	12.5	"
	4	120	96.7	75.7	41.7	33.7	13.0	"
	5	120	98.3	93.0	41.0	38.3	13.5	"
7	1	110	90.0	67.7	44.3	32.7	13.5	"
	2	120	93.0	77.0	41.0	40.3	13.5	"
	3	110	83.3	71.3	39.3	33.7	13.0	"
	4	120	94.0	80.7	43.7	39.3	14.0	"
	5	115	87.3	77.3	46.0	40.0	13.5	"
8	1	110	87.3	79.3	47.7	44.3	14.0	"
	2	140	91.3	81.0	48.7	40.0	13.5	"
	3	115	88.7	78.7	43.7	41.3	14.0	"
	4	125	90.3	83.3	46.0	41.0	13.5	"
	5	120	89.0	75.0	47.7	37.0	13.5	"
9	1	120	89.0	79.7	45.0	38.7	13.5	"
	2	130	94.3	85.0	47.0	40.0	13.0	"
	3	130	90.3	80.3	47.0	39.3	13.5	"
	4	130	88.0	77.7	44.3	41.0	13.0	"
	5	135	96.6	82.0	47.7	40.0	13.5	"

Table 42. (Continued)

M14 x M1984								
Plant number	Spikelet number	Glume length	Mean anther length		Mean anther width		Mean diameter of microspore or pollen	Stage of development of microspore or pollen
10	1	120	91.7	80.7	48.7	38.7	13.5	99
	2	115	92.0	80.7	48.0	32.7	13.5	"
	3	120	94.7	79.0	48.7	37.7	13.5	"
	4	130	93.3	79.3	47.0	36.0	13.5	"
	5	110	89.0	78.7	43.7	35.3	13.5	"
11	1	130	91.0	81.7	47.0	38.7	13.5	"
	2	130	94.0	81.7	46.0	41.0	13.5	"
	3	120	83.0	68.7	44.3	40.0	11.0	"
	4	135	94.0	81.0	46.0	38.7	13.5	"
	5	125	93.3	80.3	46.0	38.7	13.5	"
12	1	120	91.0	80.3	45.3	36.3	12.5	"
	2	120	89.0	72.7	46.0	39.3	12.5	"
	3	120	90.3	81.7	45.0	39.3	12.5	"
	4	130	89.3	78.7	47.0	41.0	12.5	"
	5	120	87.3	78.3	47.0	41.0	12.5	"
13	1	130	99.3	80.3	50.0	42.7	14.0	"
	2	120	96.3	81.3	48.7	42.0	14.5	"
	3	120	100.0	83.3	50.0	37.0	14.5	"
	4	125	101.7	81.7	49.3	42.0	14.5	"
	5	120	100.7	83.0	47.7	37.7	14.5	"